PCT WORLD INTELLECTUAL PROPERTY ORGANIZATION
International Bureau

INTERNATIONAL APPLICATION PUBLISHED UNDER THE PATENT COOPERATION TREATY (PCT)

(51) International Patent Classification 6 : A61K 38/00 — A1

(11) International Publication Number: **WO 99/43338**

(43) International Publication Date: 2 September 1999 (02.09.99)

(21) International Application Number: PCT/US99/04351

(22) International Filing Date: 26 February 1999 (26.02.99)

(30) Priority Data:
09/031,122 26 February 1998 (26.02.98) US

(63) Related by Continuation (CON) or Continuation-in-Part (CIP) to Earlier Application
US 09/031,122 (CIP)
Filed on 26 February 1998 (26.02.98)

(71)(72) Applicant and Inventor: GLINSKII, Guennadi, V. [UA/US]; Metastat, Inc., 10835 Altman Row, San Diego, CA 92121 (US).

(74) Agent: SHUSTER, Michael, J.; McCutchen, Doyle, Brown & Enersen, LLP, Three Embarcadero Center, San Francisco, CA 94111 (US).

(81) Designated States: AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, CA, CH, CN, CU, CZ, DE, DK, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MD, MG, MK, MN, MW, MX, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, UA, UG, US, UZ, VN, YU, ZW, ARIPO patent (GH, GM, KE, LS, MW, SD, SL, SZ, UG, ZW), Eurasian patent (AM, AZ, BY, KG, KZ, MD, RU, TJ, TM), European patent (AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE), OAPI patent (BF, BJ, CF, CG, CI, CM, GA, GN, GW, ML, MR, NE, SN, TD, TG).

Published
With international search report.
Before the expiration of the time limit for amending the claims and to be republished in the event of the receipt of amendments.

(54) Title: ANTIBACTERIAL DRUGS, METHODS FOR THEIR DESIGN AND METHODS FOR THEIR USE

(57) Abstract

Antibacterial drugs and general methods of design and use of the inhibitors of bacterial growth and anti–bacterial drugs are described. The methods are based on design and application of the compounds blocking the assembly and function of DNA–dependent RNA–polymerase (RNAP) by targeting protein–protein contact site(s) and nucleic acid binding site(s) of at least one protein component of RNAP and thereby inhibiting the subunit–subunit interactions essential for RNAP assembly and function. Specific examples of the antibacterial drugs, proposed method of anti–bacterial drug design, and use based on the inhibition of protein–protein and protein nucleic acid interactions are presented.

FOR THE PURPOSES OF INFORMATION ONLY

Codes used to identify States party to the PCT on the front pages of pamphlets publishing international applications under the PCT.

AL	Albania	**ES**	Spain	**LS**	Lesotho	**SI**	Slovenia
AM	Armenia	**FI**	Finland	**LT**	Lithuania	**SK**	Slovakia
AT	Austria	**FR**	France	**LU**	Luxembourg	**SN**	Senegal
AU	Australia	**GA**	Gabon	**LV**	Latvia	**SZ**	Swaziland
AZ	Azerbaijan	**GB**	United Kingdom	**MC**	Monaco	**TD**	Chad
BA	Bosnia and Herzegovina	**GE**	Georgia	**MD**	Republic of Moldova	**TG**	Togo
BB	Barbados	**GH**	Ghana	**MG**	Madagascar	**TJ**	Tajikistan
BE	Belgium	**GN**	Guinea	**MK**	The former Yugoslav Republic of Macedonia	**TM**	Turkmenistan
BF	Burkina Faso	**GR**	Greece			**TR**	Turkey
BG	Bulgaria	**HU**	Hungary	**ML**	Mali	**TT**	Trinidad and Tobago
BJ	Benin	**IE**	Ireland	**MN**	Mongolia	**UA**	Ukraine
BR	Brazil	**IL**	Israel	**MR**	Mauritania	**UG**	Uganda
BY	Belarus	**IS**	Iceland	**MW**	Malawi	**US**	United States of America
CA	Canada	**IT**	Italy	**MX**	Mexico	**UZ**	Uzbekistan
CF	Central African Republic	**JP**	Japan	**NE**	Niger	**VN**	Viet Nam
CG	Congo	**KE**	Kenya	**NL**	Netherlands	**YU**	Yugoslavia
CH	Switzerland	**KG**	Kyrgyzstan	**NO**	Norway	**ZW**	Zimbabwe
CI	Côte d'Ivoire	**KP**	Democratic People's Republic of Korea	**NZ**	New Zealand		
CM	Cameroon			**PL**	Poland		
CN	China	**KR**	Republic of Korea	**PT**	Portugal		
CU	Cuba	**KZ**	Kazakstan	**RO**	Romania		
CZ	Czech Republic	**LC**	Saint Lucia	**RU**	Russian Federation		
DE	Germany	**LI**	Liechtenstein	**SD**	Sudan		
DK	Denmark	**LK**	Sri Lanka	**SE**	Sweden		
EE	Estonia	**LR**	Liberia	**SG**	Singapore		

ANTIBACTERIAL DRUGS, METHODS FOR THEIR DESIGN AND METHODS FOR THEIR USE

I. INTRODUCTION

A. Technical Field

This invention relates to the field of inhibitors of protein function, particularly proteins that regulate bacterial growth.

B. Background

Development of resistance to anti-bacterial drugs represents one of the most challenging problems in the treatment of infections. Bacterial cells are capable of escaping from the strike of the most sophisticated weapons simply by changing the structures which are the targets of drugs via mutations in corresponding genes. The "golden bullet" of one moment cannot destroy effectively the mutated bacteria of the next moment. Furthermore, because of continuing drug application, the mutant cells gain a selective growth advantage and take over in the bacterial population, replacing parental drug-sensitive cells with a drug-resistant bacterial strain.

Antibiotic resistant bacterial strains cause major problems in current medical practice. Neu (1993). Since the introduction of nalidixic acid in 1970, no new chemical class of antibiotic has been introduced into medicine in the past two decades. Silver and Bostian (1990). Main experimental approaches have focused on modification of the structures of existing antibiotics via chemical manipulation to develop novel derivatives with an improved spectra of antibacterial activities. However, because the mechanism of biological activity of the antibiotic derivatives remains the same, resistance to such antibiotics usually has emerged rapidly.

Therefore an anti-bacterial drug and a method of treating bacterial infections which discourages the development of resistance is needed. An effective novel antibiotic in the antibiotic resistance era would be an agent that has a new chemical structure and novel mechanism of antibacterial activity. Consequently, it

should not suffer from existing intrinsic or induced antibiotic resistance pathways, and ideally it should directly target a common resistance mechanism. Hancock (1997). The key structural components of bacterial cells such as membranes and/or enzymes essential for bacterial cell survival comprising multi-subunit complexes are particularly attractive targets of antibacterial drug design.

One possible molecular target of an anti-bacterial drug that satisfies these criteria is RNA-polymerase. The transcription of genes is an essential process of the life cycle of monocellular as well as multicellular organisms. DNA-dependent RNA-polymerase (RNAP) is the key enzyme performing gene transcription. RNAP is a multiprotein complex consisting of five subunits: two α-subunits (each 36.5 kDa), one β-subunit (150.6 kDa), one β'-subunit (155.2 kDa), and one σ-subunit. The RNAP α subunit serves as the initiator of the assembly of RNAP according to the following sequence (Zillig *et al.* (1976); Ishihama (1981)):

$$\alpha + \alpha = \alpha 2;\ \alpha 2 + \beta = \alpha 2\beta;\ \alpha 2\beta + \beta' = \alpha 2\beta\beta';\ \alpha 2\beta\beta' + \sigma = \alpha 2\beta\beta'\sigma.$$

During this process an essential catalytic RNAP core enzyme, $\alpha 2\beta\beta'$, interacts with one additional regulatory subunit called σ-factor to form the transcriptionally competent RNAP holoenzyme $\alpha 2\beta\beta'\sigma$. Bacterial cells have many regulatory σ-subunits comprising a family of structurally related proteins. Helmann and Chamberlin (1988) and Lonetto *et al.* (1992). Each promoter recognition σ-subunit is targeting RNAP to a specific subset of genes, transcription of which is necessary at any given period of bacterial life cycle or growth conditions. Thus, one of the multiple species of σ-subunits binds to the RNAP core enzyme and forms a unique σ-specific RNAP holoenzyme. Helmann and Chamberlin (1988) and Ishihama (1993). This σ-dependent functional differentiation of RNAP core enzyme into RNAP holoenzyme is the most efficient mechanism of alteration of the promoter recognition property of RNAP and transcriptional regulation of a specific set of genes in bacteria. Helmann and Chamberlin (1988) and Ishihama (1988).

II. SUMMARY OF THE INVENTION

The invention provides antibacterial compounds, methods for the design of antibacterial compounds and methods for their use which block the assembly of a multi-subunit complex, preferably RNAP, thereby interfering with bacterial life cycle. More specifically the compounds are designed to inhibit the subunit-subunit interactions and assembly necessary for enzyme function.

The invention provides antibacterial drugs that comprise a compound that blocks the binding of at least one protein subunit of the multi-subunit complex to another protein subunit of the complex or targets nucleic acid binding sites of the complex. The compound may prevent any subunit in the multi-subunit enzyme from binding, but preferably the compound targets RNAP. The compound preferably targets the binding sites of the σ-subunit and β′-subunit of RNAP. More preferably, the compound targets amino acid residues 60 to 135 of the β′-subunit. Specific exemplary compounds are described.

The invention provides methods of interfering with bacterial life cycle, inhibiting bacterial growth, killing bacterial cells, and treating infection comprising bringing bacterial cells into contact with a compound that blocks the binding of at least one protein subunit of the multi-subunit complex to another protein subunit of the complex or targets nucleic acid binding sites of the complex. The method may prevent any subunit in the multiprotein enzyme from binding, but preferably the compound targets RNAP. The compound preferably targets the binding sites of the σ-subunit and β′-subunit of RNAP. More preferably, the compound targets amino acid residues 60 to 135 of the β′-subunit. Specific useful compounds for use in the method of the present invention are described.

The invention also provides methods for the design of inhibitors of bacterial growth and anti-bacterial drugs. The method may include the following steps:

a) identifying a region of a enzyme subunit that is involved in making subunit-subunit contacts;

b) performing a fine mapping of the region;

c) designing a compound that binds to the region;

d) developing of inhibitory drug molecules; and

e) testing the effect of the drug molecules on bacterial growth and infections.

The region may be in any subunit in the multiprotein enzyme, but preferably the region is located in the binding site of the σ-subunit and β′-subunit of RNAP. More preferably, the region includes amino acid residues 60 to 135 of the β′-subunit.

III. BRIEF DESCRIPTION OF THE DRAWINGS

FIG. 1 illustrates the identification of the σ-binding region on the β′ subunit of RNAP. The heavy bar represents the β′ polypeptide with evolutionarily conserved regions designated by capital letters. Puhler *et al.* (1989) and Sweetser, *et al.* (1987).

FIG. 2 shows the results of two competition enzyme linked immunoadsorption assays (ELISA) testing β′ insert-containing virions for their ability to bind bio-σ^{38} protein in a competition ELISA using a molar excess of a cold σ^{38} protein as a competitor.

FIG. 3 shows five chemically synthesized peptides and their relation to the β′ subunit of RNAP.

FIG. 4 shows binding of synthetic L-peptides derived from the σ-binding region of the β′ subunit of RNAP to purified σ^{38} in a direct ELISA.

FIG. 5 shows inhibition of bacterial growth by synthetic L-peptides derived from the σ-binding region of the β′ subunit of RNAP.

FIG. 6 shows binding of synthetic β′-derived peptides to σ^{70}.

FIG. 7 shows competition of purified σ^{70} and σ^{38} proteins for peptide 4 binding to σ^{38}-MBP fusion protein.

IV. DEFINITIONS

RNAP means ribonucleic acid (RNA) polymerase. Unless otherwise specifically stated RNAP refers to the RNAP holoenzyme.

Peptide derivatives means peptides produced by changes in the primary peptide such as acetylation, amidation, methylation, amino acid substitution, insertion of D-amino acids, decarboxylation, oxidation, analogs and fragments of the primary peptide, and peptides which differ from the primary peptide by the identity of one or more amino acid residues, for example, deletion, substitution and addition analogs. Peptide derivatives share some or all of the properties of the primary peptide.

V. DESCRIPTION OF THE SPECIFIC EMBODIMENTS

One of the objectives of the present invention is to develop a method of antibacterial drug design that would overcome one of the general mechanisms of the development of drug resistance by bacterial cells. The following broad criteria for selection of potential molecular targets for antibacterial drug design should be considered: 1) the identified molecular target should comprise a multi-subunit complex; 2) subunit-subunit interactions and binding should be required for its assembly and function; 3) the function of the target molecules should be essential for bacterial cell survival.

One of the molecular targets that satisfies these criteria is bacterial RNA polymerase. The methods provided by this invention target RNA polymerase, an enzyme essential for bacterial cell survival. The invention can be applied to any other multiprotein complex or enzyme necessary for bacterial growth or survival, for

example, enzymes that affect how bacteria hydrolyze drugs or pump drugs out. The invention overcomes the general mechanism of drug resistance development by bacterial cells by targeting subunit-subunit contact sites of the enzyme. The compounds bind to a subunit-subunit contact site on at least one subunit of the enzyme, thereby inhibiting the subunit-subunit interactions and blocking enzyme assembly and function. In this case, a single mutation that would cause a modification to the subunit-subunit contact site, and therefore resistance to the effects of the compound, would be lethal to the bacteria. Any modification of one side of the contact site would alter the subunit-subunit recognition interactions required for assembly and function of the enzyme. Development of resistance to this type of anti-bacterial compound would require simultaneous mutations in the binding sites of at least two interacting subunits of the enzyme. In other words, complementary mutations in both sides of a binding site would be required in order to preserve the recognition essential for enzyme assembly. In addition, the present invention may reverse the existing drug resistance mechanism used by bacteria by blocking transcription of the pump that expels existing antibiotics from the bacterial cells.

The invention provides antibacterial drugs which block the assembly of a multi-subunit complex, prefereably RNAP, thereby interfering with bacterial life cycle, methods for their design, and methods for their use. More specifically the compounds are designed to inhibit the subunit-subunit interactions and assembly necessary for enzyme function or target nucleic acid binding sites of the complex. A method of designing the antibacterial drug provided by the invention may include the following steps:

a) identifying a region of a enzyme subunit that is involved in making subunit-subunit contacts;

b) performing a fine mapping of the region;

c) designing a compound that binds to the region;

d) developing inhibitory drug molecules; and

e) testing the effect of the drug molecules on bacterial growth and infections.

A. Identifying the Regions of the Subunits Involved in Assembly

Any region involved in assembly of RNAP may be targeted for the purposes of the present invention. For example, protein-protein interactions between α- and β- subunits is an early event in RNAP assembly that is absolutely essential for assembly of the core enzyme. Therefore, interference with RNAP assembly by targeting α-β, α-β′ or β-β′ contact sites will completely abolish formation of RNAP core enzyme at the stage of α2β tertiary or α2ββ′ tertiary complex formation and block gene transcription. Such interference could be achieved by targeting either the β-binding site on the α-subunit or the α-binding site on the β-subunit.

Alternatively, the β′-σ subassembly region can be targeted. During the last step of RNAP assembly an essential catalytic RNAP core enzyme comprising a tertiary complex α2ββ′ interacts with one additional regulatory subunit called σ-factor to form the transcriptionally competent RNAP holoenzyme α2ββ′σ. Blocking of σ-core binding would interfere with RNAP holoenzyme formation, render gene transcription impossible, and kill bacterial cells. Since σ-core binding is facilitated by β′-σ interactions, interference with RNAP assembly at this step is accomplished by targeting either σ-binding site on β′-subunit or β′-binding site on σ-subunit.

In addition, to regions involved in the assembly of RNAP, regions involved in RNAP-nucleic acid interactions in the transcription elongation complex may be targeted in the present invention. The current model of transcription elongation implies that RNAP holds nucleic acids, DNA and RNA, in an extremely stable tertiary complex while allowing both RNA and DNA molecules to pass through the RNAP molecule. Nudler, *et al.* (1998). A snap shot of the tertiary elongation complex reveals three contiguous structural-function sites that constitute a single unit within the RNAP molecule: a) The double-stranded DNA-binding site ahead of the transcription bubble, b) the RNA-DNA heteroduplex-binding site, and c) the

upstream RNA-binding site. Nudler, *et al.* (1998). The same highly evolutionarily conserved protein regions of RNAP participate in binding of both DNA and RNA in the transcription elongation complex indicating that DNA entry and RNA exit may occur close together in the RNAP molecule. Nudler, *et al.* (1998). The nucleic acid-binding regions of RNAP are responsible for the unique feature of RNAP as a strong DNA-binding protein with no affinity for particular DNA sites. Therefore, these unique nucleic acid-binding protein regions of RNAP are critical for maintaining the integrity and continuity of the transcription elongation process and ensuring the processivity of RNAP. Nudler, *et al.* (1996). Targeting the nucleic acid-binding sites of RNAP with specific small molecules inhibitor will be a very efficient way of blocking gene transcription and killing bacterial cells. Both the β-subunit and the β′-subunit have been implicated in interactions with DNA and RNA within the transcription elongation complex. Nudler, *et al.* (1996) and Nudler, *et al.* (1998).

Several experimental approaches could be employed in order to identify the protein-protein contact regions involved in the subunit-subunit interactions during RNAP assembly. Among the approaches available to identify the regions are:

1) Molecular mutagenesis study and polymerase chain reaction (PCR)-based subcloning of the fragments of interacting proteins. Mutations in the gene encoding protein of interest, isolation of truncated and/or mutated versions of the protein and subsequent analysis of their binding properties utilizing ELISA, immunoprecipitation, bioactivity assays, or chemical cross-linking assays represent typical experimental steps when this approach is employed. Coggins, *et al.* (1977); Hillel, *et al.* (1977); McMahan, *et al.* (1994); Glass, *et al.* (1986); Siegel, *et al.* (1989); Lesley, *et al.* (1989); Lesley, *et al.* (1991); Fukuda, *et al.* (1974); and Luo, *et al.* (1996).

2) There are two methods of application of small metal chelates that cleave polypeptide chains at sites determined by proximity to the chelate. One of these methods applies untethered chelates for random protein cleavage. Greiner, *et al.*

(1996). The second method applies chelates tethered at specific protein site(s) for site-directed protein cleavage. Owens, *et al.* (1998). The appearance of the cleavage sites in the absence of one subunit bound to the multi-subunit complex serves as a guide to the potential protein-protein binding sites when nontethered chelates are used. On the other hand, the appearance of the cut sites in the presence of the subunit with tethered chelates serves as an indicator of the possible protein-protein binding sites when tehtered chelates are applied.

3) Limited proteolysis of the corresponding protein-protein complexes and subsequent analysis of the bound peptide fragments in order to define protein domains participating in protein-protein complex formation. Wang et al. (1997). Parts of the interacting proteins that are directly involved in protein-protein interactions during complex formation are usually less susceptible for proteolytic degradation and are protected during limited proteolysis. Therefore, the bound peptide fragments of the interacting proteins, which are protected from degradation, can be isolated and characterized. Alternatively, proteins of interest could be fragmented into several small peptide fragments. These peptide fragments could be isolated, and their binding properties characterized.

4) Hydroxyl-radical protein footprinting of the protein-protein contact sites. Heyduk, *et al.* (1996) and Wang, *et al.* (1997). According to this method, initially the complex of proteins of interest should be purified to homogeneity. Then hydroxyl-radical-mediated cleavage of the complex is performed and the cleavage pattern compared quantitatively with the cleavage pattern of each protein alone. The assumption is that the regions of the proteins that are protected from cleavage when the complex is formed are directly involved in protein-protein binding and form the protein-protein contact site.

5) A combinatorial chemistry approach utilizing random peptide bacteriophage display libraries for initial screening for peptide sequences with high binding ability toward corresponding target proteins (Scott and Smith (1990); Devlin, *et al.*, (1990); Cwirla, *et al.*, (1990)) and subsequently applying this information for

identification of the similar peptide sequences in the primary structures of interacting proteins. The phage display method is based on repetitive synthesis and rescreening of peptides with desired binding specificity toward selected target molecules. The repetitive amplification of peptides interacting with target molecules in subsequent rounds of selection typically leads to the isolation and identification of specific peptide binders from a large random pool of peptide sequences displayed on phage surfaces. Subsequently synthetic analogues of these specific peptide binders could be used to target proteins of interest. Alternatively, the sequence homology search could be performed for identification of similar peptide sequences in the proteins that are known to interact with and bind to the target protein during multi-subunit complex formation.

Any of these procedures can be used alone or in combination to identify the protein-protein contact regions involved in the subunit-subunit interactions.

B. Fine Mapping

The step of fine mapping of the protein-protein contact site is defined in the present invention as the identification of a minimal binding fragment of the protein that is directly involved in subunit-subunit interactions and binding and that can be chemically synthesized. Overlapping peptides within the binding region are synthesized. The peptides are then tested for binding ability and compared with other peptides to determin the peptide with the best binding properties.

C. Designing a Compound that Binds to the Region

Any of the above described approaches or their combination could be useful in the design of the potential inhibitory molecules. Particularly useful should be the application of the combinatorial chemistry approach using either a genetically encoded library of molecules or synthetic small molecule libraries. Identified peptide sequences of selected RNAP subunits that represent the corresponding subunit-subunit contact sites may themselves serve as inhibitors of RNAP assembly and

bacterial growth. For example, several derivatives or fragments of the identified peptide sequences could be useful as antibacterial drugs. Such derivatives may be developed by way of introduction of one of the many possible chemical modifications of identified peptide sequences such as acetylation, amidation, methylation, amino acid substitution, insertion of D-amino acid, decarboxylation, oxidation, etc. Fragments of identified peptide sequences could be chemically synthesized and tested for antibacterial activity. Structural information obtained by NMR and X-ray analysis of identified peptides could be applied for design of non-peptide synthetic analogues with desired binding specificity and antibacterial activity.

Alternatively, identified peptide sequences of selected RNAP subunits that represent the corresponding subunit-subunit contact sites may serve as a molecular target for design of specific inhibitory compounds that would bind to the above named peptide sequences on corresponding RNAP subunits thereby preventing RNAP assembly and function. Several well known approaches could be utilized by those of ordinary skill in the art for identification and design of specific inhibitory compounds targeting particular peptide sequences. Identification of specific molecules with desired binding specificity toward identified peptide sequences could be performed employing combinatorial chemistry approaches such as application of genetically encoded libraries, namely phage display (Scott and Smith, (1990); Devlin, *et al.* (1990); Cwirla, *et al.* (1990)), "peptide on plasmid" (Cull, *et al.* (1992)), and *in vitro* translation-based systems (Mattheakis, *et al.* (1994)), as well as synthetic small molecule libraries (Bunin, *et al.* (1994); Gordon, *et al.* (1994); Dooley, *et al.* (1994)).

D. Developing Inhibitory Small Drug Molecules.

Unfortunately, peptides, particularaly L-peptides, have a number of deficiencies as drug candidates. Some of the major problems with the peptides as drug candidates are: 1) the possibility of development of immunogenic reactions; 2) sensitivity to proteolysis and instability in the digestive system; and 3) potential difficulties in delivery into bacterial cells due to restrictions for transport across the cell membrane. If the peptides developed in the previous step are unsuitable for

application as drugs, it will be necessary to transform the active peptide prototype molecules into active peptidomimetic drug candidates. Libraries of small organic molecules, which are inherently better prospective drug candidates, developed using a variety of organic building blocks and reactions have been developed. See, for example, Hogan (1996, 1997). As described below, combinatorial chemistry using these libraries in conjunction with a computational approach for structure-guided drug design is used to develop the peptideomimetic drug candidates.

There are two major types of combinatorial libraries of small organic molecules that may be used during drug discovery and development: random combinatorial libraries and focused or targeted combinatorial libraries. The huge random combinatorial libraries of small organic molecules are designed for hit discovery. High throughput screening technology is used to identify lead compounds. The focused or targeted combinatorial libraries of small organic molecules are designed from optimization of lead compounds to identify viable drug candidates. The design and synthesis strategies as well as size and diversity requirements are different for these two types of libraries. Hogan (1996, 1997) The present invention uses both types of libraries in combination to develop drug prototypes.

1. High throughput screening for antibacterial drug candidates using combinatorial libraries of small organic molecules.

Peptide-protein binding ELISA is used for high throughput screening of commercially available random combinatorial libraries of small organic molecules with synthetic biotinilated L-peptides comprising the minimal binding fragments within subunit-subunit contact sites. A hit discovery rate for screening each small molecule combinatorial library would be established. The hit discovery rate is used as a quantitative comparison between the different combinatorial libraries. The anticipated hit discovery rate for high throughput screening of random combinatorial libraries should be from 0.1 to 1.0%. The expected hit discovery rate for screening targeted combinatorial libraries designed by application of the computational

approach described below is from 1.0 to 10%. Different libraries should be screened (or developed and screened using the computational approach described below) until a hit discovery rate of approximately 10% with Ki<10nM in peptide protein binding ELISA is acheived.

Several different existing libraries that have been useful in finding other drugs that mimic peptide features would be useful for screening including:

i) Benzodiazepin core-based libraries (Evans, *et al.* (1988); Bunin & Ellman (1992); Hobbd De Witt, *et al.* (1993));

ii) Libraries of hydroxyaminimides comprising 1,000 compounds synthesized from 30 building blocks (Peisach, *et al.*, (1995); Hogan (1996); Hogan (1997));

iii) α-Ketoamide-based libraries consisting of 1,600 compounds derived from 38 building blocks (Baldino, *et al.* (1997); Hogan (1996); Hogan (1997));

iv) Libraries of arylidine diamides comprising 15,000 compounds derived from less than 100 reagents (Hogan (1997));

v) Libraries of thiazolidines (Patek, *et al.* (1995)) and hydantoins (Hobbd De Witt, *et al.* (1993)).

Preferably, the high throughput screening protocol is performed in a 96-well plate format and comprises a competition peptide-protein binding ELISA in which the individual compounds from a given library will compete for binding of biotinilated peptide ligand with a receptor protein (σ-, β′-, or α-subunit of RNAP). Preferably, the cut-off level is established at Ki<50 nM for initial screening of random combinatorial libraries.

2. General description of the computational approach for structure-guided antibacterial drug design.

A higher hit discovery rate may be achieved through the combination of screening combinatorial libraries and using the computational approach to drug design in order to develop targeted libraries. The computational approach may be employed for translation of the structural information derived from the sequences comprising the minimal subunit-subunit binding fragments into small organic molecule antibacterial drug candidates, which then may be tested for efficacy or may be added to a targeted library of compounds for further screening. The following steps may be used:

Homology modeling. Homology modeling of a binding region is used to determine conformational information on the region and to establish the 3D positions within the binding region of the amino acids essential for binding the corresponding subunit. Homology modeling may be performed using various commercially available software, such as the Insight II program (Molecular Simulations, Inc., San Diego, CA) and the Insight II-Homology module.

Building of an active site map. An active site map may be derived from the homology model using commercially available software such as Cerius2 Structure-Based Focusing module software (Molecular Simulations, Inc., San Diego, CA). Structural-functional information on the minimal binding fragment is used for editing of the active site map to dissect essential query features that are translated into 3D pharmacophores.

Virtual screening. This step comprises virtual screening of a database of commercially available compounds (ACD) against 3D pharmacophores derived in the previous step with commercially available software such as Catalyst, HipHop, or Ludi, all by Molecular Simulations, San Diego, CA.

***De novo* drug design.** These techniques use 3D searching of large databases to identify small molecule fragments which can interact with specific sites in the receptor, bridging fragments with the correct size and geometry, or framework structures which can support functional groups at favorable orientations. *De novo* design approaches should ultimately lead to development of the targeted combinatorial libraries of small molecules and to selection of the targeted sub-libraries of drug candidates from random libraries by searching a large database of commercially available small organic molecule libraries. Various software tools, such as Catalyst, HipHop, and Ludi (Molecular Simulations, Inc., San Diego, CA), can be used in *de novo* design. Ludi software is a powerful tool for *de novo* rational drug design. A software tool, such as the Ludi program, is used to fit molecules into the active site of a receptor by identifying and matching complementary polar and hydrophobic groups. An empirical scoring function is used to prioritize the hits. Ludi also can suggest modifications that may increase the binding affinity between an existing ligand and the receptor. Ludi/ACD links the design tools of Ludi to MDL's Available Chemicals Directory. Ludi/ACD provides access to over 65,000 commercially available structures to accelerate the search for drug candidates. For implementation of the *de novo* design program, both a database search as well as a shape search and alignment are used.

3. Combining combinatorial library screening and the computational approach.

The analog-based drug design represents the most common and traditional application of computational tools to rational drug design. Starting with a collection of peptide molecules of known structure and activity, one may develop a 3D pharmacophore hypothesis as well as a quantitative structure-activity model (QSAR), that is converted into a search query (pharmacophore query) or a predictive molecular formula. One the uses the pharmacophore query to search a 3D database for structures that fit the hypothesis within a certain tolerance, or use the QSAR model to predict activities on novel compounds. Development of a predictive model would allow formulation of new active compounds that possess better overall therapeutic

profiles, for example compounds that will be more stable, selective or orally active. The binding assay data will be generated from a survey of a series of commercially available compounds that will determine which compound binds to a particular peptide receptor mimic with the Ki<50 nM at the initial screening stage. These data are used to develop a summary of the 3D interactions that might be responsible for the receptor-binding activity. After possible models are identified, each "hypothesis" can be tested in a peptide protein ELISA assay to determine which of the many models might be correct. Potentially active compounds can be selected by searching databases of known compounds that can be purchased from suppliers such as Sigma, Aldrich, or Fluka, or are available in a database of combinatorial libraries of small organic molecules from suppliers such as MSI, ArQuele, or Pharmacopia. Selected compounds subsequently are tested in peptide-protein binding ELISA to determine which compounds possess strongest activity. HipHop software (Molecular Simulations, Inc., San Diego, CA) can be used to perform a feature-based alignment of a collection of compounds onto a pharmacophore hypothesis. HipHop is used to match features, such as surface-accessible hydrophobes, charged or ionizable groups, or surface-accessible hydrogen bond donors or acceptors, against candidate molecules or searches of 3D databases.

In addition, 3D database searching techniques are some of the most successful ways to identify new structural templates from which new drug compounds can be synthesized. The pharmacophore definitions are an important key to successful 3D searching. The closer the definitions encapsulate drug-receptor interactions, such as hydrophobic areas and hydrogen bonding, the better the diversity of identified templates. Software, such as Catalyst/INFO (MSI, San Diego, CA), identifies structurally diverse leads by using a hypothesis as a search query against one or more databases that may contain up to hundreds of thousands of molecules. The Catalyst/INFO program also builds and administers databases of 3D structures from project data. Catalyst/INFO hit lists are used to select compounds for assay as well as to guide synthesis of new compounds (design of the targeted combinatorial libraries of small molecules).

Shape complementarity is one of the most important considerations in structure-based drug design. Catalyst/SHAPE software (Molecular Simulations, Inc., San Diego, CA) takes a specified 3D conformation and identifies compounds that possess similar 3D shapes. This module complements the powerful pharmacophore and hypothesis-based searching also available within the Catalyst program. One of the benefits of searching for shape similarity is to produce a more diverse and extensive list of drug candidates than traditional search methods. Consequently, application of the Catalyst/SHAPE program should maximize the chance for success by maintaining a broad list of potential drug candidates throughout the drug development process.

Since drug molecules must fit into a receptor cavity with a specific shape before they can bind, Catalyst/SHAPE provides an initial, fast selection process for potential pharmaceutical compounds by taking an input molecule and performing a 3D search and retrieving compounds that have similar shapes to the input molecule. Catalyst/SHAPE further screens 3D search results and focuses on the best drug candidates from a database of small molecule combinatorial libraries. Subsequently, Catalyst/SHAPE imports shape descriptors of the original query molecule and the matching database search results into C2.Receptor program (MSI, San Diego, CA). The C2.Receptor program provides a 3D visual environment for structural analysis. Shape similarity of the hits will be analyzed and manipulated to minimize each hit within the receptor surface model created by C2.Receptor. This analysis allow flexible fitting of the molecules into a receptor cavity.

In addition to database searches, shape searches and shape-based alignment, feature-based, or pharmacophoric, alignment approaches also are useful. Alignments based upon general chemical features such as hydrophobic areas, hydrogen bonding groups, and ionizable groups, have been shown to produce one of the most reliable and robust inputs for 3D-QSAR techniques. Their features and 3D positions provide ideal 3D queries with which to search structural databases. The HipHop program (Molecular Simulations, Inc., San Diego, CA) can be used for this purpose. HipHop performs feature-based alignment of a collection of compounds onto a pharmacophoric hypothesis. HipHop is used to match features, such as

surface-accessible hydrophobes, charged or ionizable groups, or surface-accessible hydrogen bond donors or acceptors, against candidate molecules or searches of commercially available 3D databases.

Any hits identified by the random screening, can be developed as 3D pharmacophore models using Catalyst technology (Molecular Simulations, San Diego, CA). Subsequently, virtual screening of a database of commercially available compounds (ACD) against the Catalyst pharmacophore models can be performed. Approximately 100 small molecule drug candidates with the most promising potential binding ability and structural features should be identified (targeted sub-libraries of small molecule drug prototypes). The selected compounds are tested experimentally employing peptide-protein binding ELISA in order to validate the proposed 3D pharmacophore models, validate the selected structure-based design approach, and identify the essential structural building blocks for design and synthesis of the targeted small molecule combinatorial libraries of antibacterial drug candidates. Both experimental and computational methods will be employed for follow-up experiments. The hit discovery rate at this stage should be at least ten fold higher than the hit discovery rate of non-targeted libraries. These steps should be repeated until a targeted library of small organic molecules is establish which yields a 10% hit discovery rate at Ki<10 nM in peptide protein binding ELISA. In addition to the higher hit discovery rate, the affinity of the ligands identified from a targeted combinatorial libraries should be higher with the Ki<10 nM when compared with Ki<50 nM for ligands identified from the random library screening.

The computational approach can be applied using L-peptide sequences that have been identified as receptors, L- and D- peptides that are prototype ligands for analog based drug design, or complementary L-peptide/D-peptide pairs as a mimic of the receptor/ligand complex. L-peptide sequences comprising the minimal subunit-subunit binding fragments can serve as receptor prototypes for design of a small molecule antibacterial drug. Structural information about the receptor may be utilized to identify new leads that can interact with the receptor active site. *De novo* design techniques can be used to propose new small molecule ligands, which are

complementary to the active site. Alternatively, synthetic biotinilated L-peptides will be employed for high throughput screening of combinatorial libraries of small organic molecules using peptide-protein binding ELISA.

L- and D-peptide sequences may serve as a molecular prototype of a ligand for the design of small molecule drug candidates employing the computational approach for structure-guided drug design. By using structural features of the identified L- and D-peptides as prototype active ligands, one can apply cluster analysis and 2D and 3D similarity search techniques to identify potential new small molecule leads. These methods rely on the principal that compounds, which look alike or have similar 3D-properties, are likely to have similar binding activity. A modification of this approach, known as "diversity assessment," can also be used to identify dissimilar compounds for combinatorial chemistry and high-throughput screening applications.

If the receptor/ligand complex 3D structure is not known, it is difficult to select an appropriate conformation for each compound and the selection of a common alignment for each of the selected conformers. For rigid compounds, the selection of conformers becomes much simpler, but the selection of an alignment still poses a significant problem. A significant benefit of the present invention is that, while not necessary for the invention, the receptor/ligand complex can be determined. In the present invention, if the complementary L-peptide/D-peptide pairs are known, they may be used as a mimc of the receptor/ligand complex. Then the most efficient computational approach, "structure-based drug design," can be used. Structure-based drug design relies heavily on molecular graphics and simulation technology. One can address highly specific receptor-ligand interactions using these techniques, including consideration of alternative modes of binding and conformational changes in the receptor structure. The first step in this approach is to generate a hypothesis from the structure and activity data. The next step is to search for compounds that match the hypothesis, using software such as Catalyst (Molecular Simulations, Inc., San Diego, CA). New molecules can then be designed through *de novo* design programs to fit your hypothesis. The most promising candidates can be synthesized and assayed as described above. If receptor structure is known, *de novo* design can be used to build a

receptor-surface model (a model for the receptor site) and to construct compounds inside this model that fit sterically and complement the putative receptor interactions.

E. Testing the Inhibitory Molecule's Effect on Bacterial Growth.

Testing the inhibitory effect of the potential antibacterial compounds can be performed in a variety of experimental conditions both *in vitro* and *in vivo* and usually requires the step of bringing bacterial cells into contact with the compound of interest. The bacteria can be grown in appropriate cultivation conditions depending on the metabolism of the bacteria. Time course and dose course studies can be used to determine growth or survival of the bacteria. Growth can be monitored by looking at optical density or by plating bacteria to see how many survive or are able to form colonies.

F. Methods of Using the Compounds

The compound can be used as described above and in the examples, below. In addition the compound may be administered orally or parenterally to an infected individual. The compound also may applied topically to affect local bacterial infections.

EXAMPLE 1

It has been established that during RNAP assembly the regulatory σ-subunit interacts with and binds to the β'-subunit of the core enzyme. Yura and Ishihama (1979); Ishihama (1981); Ishihama (1990); Helmann and Chamberlin (1988). Therefore, we selected the binding of the σ-subunit to the β'-subunit as an example of a potential target step in the RNAP assembly for anti-bacterial drug design.

Escherichia coli cells have two principal σ-subunits of RNAP named σ^{70} and σ^{38}. Tanaka, *et al.* (1993). σ^{70} is responsible for transcription of genes essential for growth of bacterial cells in the exponential phase, whereas σ^{38} is

important for transcriptional regulation of genes essential for survival in the stationary phase of bacterial growth. Lange and Hengge-Aronis (1991); Mulvey and Loewen (1989); Nguyen, *et al.* (1993); Tanaka, *et al.* (1993).

Identification of σ-factor binding region on β'-subunit of RNAP

A 76 amino acid region was identified by a sequence homology search comparing β' amino acid sequence and sequences of 38 peptide inserts of σ-binding virions selected through peptide phage display library screening.

Bioaffinity selection of σ^{38}-binding virions

We employed a combinatorial chemistry approach for initial identification of short peptide sequences displaying a σ-factor binding activity. The 15 and 6 amino acid random phage display libraries constructed in the fUSE vector and the bacterial strain K91Kan were utilized in these experiments. Scott and Smith (1990), and Smith and Scott (1993). The phage display libraries were first affinity selected against biotinilated σ-factor 38-maltose binding protein (σ^{38}-MBP) fusion protein using a similar affinity selection protocol for both 6-mer and 15-mer libraries.

The affinity selection procedure was based on the biopanning method described in Scott and Smith (1990), and Smith and Scott (1993). The reagents, phage libraries, and bacterial strains, as well as the experimental procedures were similar to those described in Peletskaya, *et al.* (1996) and Peletskaya, *et al.* (1997). Briefly, plastic dishes were first coated with streptavidin, washed with TPBS (phosphate buffered saline, pH 7.4, 0.5% (v/v) Tween-20), and blocked with 3% (w/v) BSA prior to addition of biotinylated antigen. Biotinylated antigen was immobilized on the streptavidin-coated dishes by two hour incubation at 20°C in TPBS buffer containing 1 mg/ml of dialyzed BSA. Immobilization of biotinylated antigen was continued further for an additional one hour in the presence of 0.1 mM biotin. The loading amount of biotinylated antigen was 14 μg and 1.4 μg in the first and second round of biopanning, respectively.

The dishes were washed six times with TPBS prior to incubation with phage. Each library that was used in the biopanning protocol, was initially pre-incubated with MBP-coated dishes in order to diminish the background of potential MBP-binding virions. Phage library was added to antigen-coated dishes and incubated overnight at 4°C in TPBS buffer containing 0.1 mM biotin. Six five-minute washings with TPBS were performed after each round of phage binding prior to acid elution of antigen-bound phage clones. Phage eluted in the preceding round of biopanning was used as an input phage in the subsequent round of affinity selection. Three sequential rounds of biopanning with increased stringency were performed in each affinity selection protocol. In the last round of biopanning three separate affinity selection procedures were performed for both libraries in which phages were pre-incubated for 30 minutes with the 100 ng, 10 ng, or 1 ng of biotinilated σ^{38}-MBP before the streptavidin capture. Eluates from the final rounds of biopanning were plated out on agar plates with 20 μg/ml tetracycline and 100 μg/ml kanamycin, yielding 1.9×10^{-4} - 2.4×10^{-3} % (% yield = eluted phage ÷ input phage x 100). Clonal phage stocks were prepared from the individual colonies.

Identification of σ^{38}-binding peptide sequences displayed on phage surface

The genes of phage DNA encoding the random peptide inserts displayed on the phage surfaces of 38 individual phage isolates from the two libraries were sequenced by a modified dideoxy sequencing methodology utilizing a ^{32}P-labeled oligonucleotide primer located 15 nucleotides downstream of the pIII gene cloning site. Haas and Smith (1993). The obtained amino acid sequences were grouped into families of conserved amino acids or motifs with the assistance of the FASTA sequence analysis program present in the GCG software package (Genetics Computer Group, Inc. Madison, WI, USA). Pearson, *et al.* (1988). 30% of the σ^{38}-binding phages selected from 15-mer phage display library screening exhibited identical σ^{38}-binding peptide sequence Arg Leu Tyr Try Val Try Phe Pro Ala Pro Val Ser Pro Ser Val Gly (SEQ ID NO:1) (RLYYVYFPAPVSPSVG), and nearly identical sequences were identified in an additional 30% of σ^{38}-binding phage clones. The only

amino acid substitutions were C at position #1 at the amino terminus in all peptides. and G or R at the position #14 in corresponding individual phage clones. Thus, 60% of σ^{38}-binding phage clones bioaffinity selected from 15-mer phage display library screening exhibited nearly identical putative σ^{38}-binding peptide sequence Arg (Cys) Leu Tyr Tyr Val Tyr Phe Pro Ala Pro Val Ser Pro Ser (Gly/Arg) Val Gly (SEQ ID NOS: 1, 10-14) (R(C)LYYVYFPAPVSPS(G/R)VG). 20% of individual phage clones selected from a 15-mer library screening displayed a highly homologous peptide insert sequence Leu Pro Arg Ser Arg Gly Ser Val His Val Leu Trp Ile Ile Ala Gly (SEQ ID NO:2) (LPRSRGSVHVLWIIAG) with a highly conserved eleven amino acid N-terminal region and a variable four amino acid C-terminal tail. The remaining 20% of the individual phage clones selected from a 15-mer library screening showed unique peptide sequences. Most of the peptide sequences derived from the 6-mer phage display library screening exhibited a high degree of similarity with 50% identity between related peptides. There are several conserved amino acid sequence motifs identified in peptide insert sequences of σ^{38}-binding virions selected from both 6-mer and 15-mer phage display library screening.

Confirmation of the σ^{38}-binding ability of selected clonally-purified phages using ELISA method

We confirmed the ability of selected clonally-purified phages to bind biotinilated σ^{38}-MBP fusion protein (bio-σ^{38}) using a sensitive peptide-displaying phage ELISA method. The most abundant 15-mer phage clone displaying Arg Leu Tyr Tyr Val Tyr Phe Pro Ala Pro Val Ser Pro Ser Val (SEQ ID NO: 15) (RLYYVYFPAPVSPSV) σ^{38}-binding peptide sequence demonstrated a significantly higher ability to capture bio-σ^{38} compared to either other σ^{38}-binding clones or the control -- a clonally purified phage clone displaying an irrelevant 15-mer peptide insert Asn Arg Ala Trp Ser Val Phe Gln Trp Gln His Ile Ala Phe Ala (SEQ ID NO:3)(NRAWSVFQWQHIAPA). It should be noted that for more stringent control of non-specific binding, the control phage clone had a peptide insert sequence containing a similar number of charged, aromatic, and hydrophobic amino acids compared to peptide inserts of the σ^{38}-binding virions. Thus, both bioaffinity

selection method and ELISA identified the phage clone displaying RLYYVYFPAPVSPSV (SEQ ID NO:15) peptide insert as an efficient binder of bio-σ^{38} protein.

Identification of the putative σ^{38}-binding region on β'-subunit of *E. coli* RNAP

Subsequently the amino acid sequences of potential σ^{38}-binding peptides were searched for homology with β'-subunit of *E. coli* RNAP employing the FASTA sequence analysis program. Several peptide sequences identified from a 6-mer phage display library screening showed at least 50% identity with amino acid sequences of the β'-subunit of RNAP. Peptide sequences selected from a 15-mer phage display library screening exhibited a 40-53% identity with the β'-subunit primary structure. Many homologous peptide sequences on the β'-subunit were clustered within a narrow highly conserved ~70 amino acid region of the N-terminal domain A of the β'-subunit of RNAP. In Fig. 1 the regions identified by a sequence homology search comparing the β' amino acid sequence and the sequences of 38 peptide inserts of σ-binding virions selected through peptide phage display library is expanded to indicate the amino acid sequence. Underlined letters indicate the amino acid residues of β' that are homologous to the amino acid sequences of peptide inserts of σ^{38}-binding virions identified through peptide phage display library screening. The homologous peptide sequences identified from a 6-mer phage display library screening were clustered within two narrow regions of N-terminal domain A of β' protein, specifically β' amino acid residues 62 to 83 (twenty two amino acids, region I) and β' amino acid residues 89 to 102 (fourteen amino acids, region II). The homologous peptide sequences identified from a 15-mer phage display library screening were clustered within the adjacent region of β' protein, specifically β' amino acid residues 114 to 132 (nineteen amino acids, region III). Fig. 1. We concluded that this region of β'-subunit of RNAP, displaying clusters of amino acid sequences homologous to the peptides selected from a phage display library screening, may be involved in β'-σ interactions during RNAP assembly and may indeed represent a sigma factor-binding site on β': Arg Ile Phe Gly Pro Val Lys Asp His Glu Cys Leu

Cys Gly Lys Tyr Lys Arg Leu Lys His Arg Gly Val Ile Cys Glu Lys Cys Gly Val Glu Val Thr Gln Thr Lys Val Arg Arg Glu Arg Met Gly His Ile Glu Leu Ala Ser Pro Thr Ala His Ile Trp Phe Leu Lys Ser Leu Pro Ser Arg Ile Gly Leu Leu Leu Asp Met Pro Leu Arg Asp Ile (SEQ ID NO:4)

(RIFGPVKDHECLCGKYKRLKHRGVICEKCGVEVTQTKVRRERMGHIELASPT AHIWFLKSLPSRIGLLLDMPLRDI, aa 60-135)

The genetically engineered viri ns displaying the 76 amino acid N-terminal region of β' subunit exhibit σ^{38}-binding in an ELISA assay

We next tested the ability to bind bio-σ^{38} protein by the genetically engineered virions displaying the 76 amino acid insert representing the putative σ^{38}-binding region of β'. The entire 76 amino acid region of β' subunit displaying sequence homology to the σ^{38}-binding peptide sequences selected from phage display library screening was PCR-amplified and cloned into the pIII gene cloning site of bacteriophage. The β' sequence-containing phages were isolated, clonally purified and sequenced to confirm the presence of the 76 amino acid β' protein insert. Subsequently, the σ^{38}-binding properties of clonally purified phages containing β' protein fragment were studied using an ELISA method. The phage clones displaying the 76 amino acid β' protein insert were immobilized on a 96-well ELISA plate and tested for their ability to capture bio-σ^{38}. The β' insert-containing virions (RNAP-1, 2, 3, and 5) exhibited a superior ability to bind bio-σ^{38} compared to the purified wild type phage (fd). Fig. 2A. The experiment was repeated with RNAP-3, which exhibited in preliminary screening the most significant binding ability toward bio-σ^{38} protein. Fig. 2B. The binding of bio-σ^{38} to the β' insert-containing phages was specific and saturable as determined in a competition ELISA assay with the x100 molar excess of cold σ^{38}. In contrast, the cold control proteins such as BSA and MBP were unable to efficiently compete for bio-σ^{38} binding to the β' insert-containing virions. Thus, we have confirmed the σ^{38}-binding ability of the 76 amino acid N-terminal region of β' subunit of RNAP. The identified σ^{38}-binding region of β' may play a physiological role in β'-σ interactions during RNAP assembly and function. It should be noted that the identified σ-binding region of β' subunit is highly conserved among different bacterial species exhibiting 80% or more homology and that known mutations in this region had a recessive lethal phenotype: the mutant plasmid failed to complement a chromosomal β' amber mutation, yet upon induction in a wild-type host, the mutant β' polypeptides did not inhibit bacterial cell growth. Nudler, *et al.* (1996).

Fine mapping of the σ-factor-binding site within β'-σ assembly region

Fine mapping of the σ-factor-binding site within the β'-σ contact region was performed by comparing the σ-binding abilities of the synthetic peptides derived from the σ-factor binding region of β'. The binding of biotin-labeled peptides to the purified σ^{38} was determined in a direct ELISA assay employing streptavidin-alkaline phosphatase reporter molecules for quantitative analysis of the bound biotinilated peptide. Purified σ^{38} (1μg per well) was immobilized overnight on a 96-well ELISA plate. The plate was blocked with 1% BSA, washed, and biotin-labeled synthetic peptides were added at the indicated concentration. After overnight incubation, the plate was washed and developed with streptavidin-alkaline phosphatase reporter according to the Sigma (St. Louis, MO, USA) protocol. Conversion of the substrate was monitored continuously in an ELISA reader at 405 nM. Non-specific binding was measured as a binding of corresponding peptides at the indicated concentration to the BSA-coated plate.

Initially we compared the σ-binding ability of the three synthetic peptides sequentially covering the potential σ-factor binding region of β' (Peptides 1, 2 and 3 on Fig. 3).

Peptide 1: Ala Arg Ile Phe Gly Pro Val Lys Asp His Glu Cys Leu Cys Gly Lys Tyr Lys Arg Leu Lys His Arg Gly (SEQ ID NO:5)(ARIFGPVKDHECLCGKYKRLKHRG)

Peptide 2: Ile Cys Glu Lys Cys Gly Val Glu Val Thr Gln Thr Lys Val Arg Arg Glu Arg Met Gly His Ile (SEQ ID NO:6) (ICEKCGVEVTQTKVRRERMGHI)

Peptide 3: Cys His Ile Trp Phe Leu Lys Ser Leu Pro Ser Arg Ile Gly Leu Leu Leu Asp Met Pro Leu Arg Asp Ile Glu (SEQ ID NO:7) (CHIWFLKSLPSRIGLLLDMPLRDIE)

As shown on Fig. 4A, peptide 1 clearly exhibited a superior binding ability compared to peptides 2 and 3. Thus, the peptide 1 sequence is likely to

represent a σ^{38} contact site on β'. Since peptide 2 showed some residual binding to the purified σ^{38}, we designed peptide 4 which covered an overlapping sub-region of peptides 1 and 2. Fig. 3.

Peptide 4: Glu Cys Leu Cys Gly Lys Tyr Lys Arg Leu Lys His Arg Gly Val Ile Cys Glu Lys Cys Gly Val (SEQ ID NO:8) (ECLCGKYKRLKHRGVICEKCGV)

Peptide 5: Cys Lys Val Arg Arg Glu Arg Met Gly His Ile Glu Leu Ala Ser Pro Thr Asa His Ile Trp Phe Leu Lys Ser Leu (SEQ ID NO:9) (CKVRRERMGHIELASPTAHIWFLKSL)

Biotinilated peptide 4 demonstrated even stronger than peptide 1 binding ability to the purified σ^{38} as determined in a direct ELISA assay. Fig. 4B. Therefore, the peptide 4 sequence of β'-subunit of RNAP is likely to represent the direct binding site for regulatory σ^{38} subunit.

In order to determine whether different σ-factors share a common binding site on the β'-subunit of RNAP, we investigated the binding ability of the σ^{70} to biotinilated peptides derived from β'-σ assembly region in a competition ELISA assay. As shown on Fig. 6, σ^{70} binding pattern to a panel of synthetic peptides is similar to that of σ^{38}: it preferentially binds biotinilated peptides 1 and 4.

We also compared the binding of two σ-subunits to biotinilated peptide 4 in a competition ELISA assay. Corresponding L-peptides derived from the σ-binding region of the β'-subunit of RNAP were chemically synthesized as described above. The peptides were labeled with biotin and tested for their ability to bind purified σ^{70} and σ^{38} in a competition ELISA assay. Purified σ^{38}-MBP fusion protein (1μg per well) was immobilized overnight on a 96-well ELISA plate. The plate was blocked with 1% BSA, washed, and biotin-labeled synthetic peptides (0.3 mg/L final concentration) were added with or without cold purified σ-subunits at 5 μM (Fig. 6) or the indicated final concentrations (Fig. 7). The maximum concentrations of sigma proteins in a competition ELISA assay were 2.5 μM. Control proteins (MBP or BSA) were added at 1 mg/ml final concentration in a 100 μl/well final volume. After

overnight incubation, the plate was washed and developed with streptavidin-alkaline phosphatase reporter according to the Sigma (St. Louis, MO, USA) protocol. Conversion of the substrate was monitored continuously in an ELISA reader at 405 nM. Non-specific binding was measured as binding of corresponding peptides at indicated concentrations to the BSA-coated plate and/or to σ38-MBP-coated plate in the of 5 μM of cold purified σ proteins. σ^{70} exhibited even higher binding ability than σ^{38} to peptide 4. Figs. 6 and 7. Therefore we concluded that the peptide 4 sequence represents part of the σ-factor binding site on the β'-subunit of RNAP.

Inhibition of bacterial growth by synthetic peptides representing a σ-factor-binding site of β'-subunit of RNAP

To directly test the notion that a fine mapping of the contact sites of RNAP subunits could lead to design of a novel class of potent anti-microbial agents, we studied the effect on bacterial growth of four synthetic peptides derived from σ-factor binding region on β'-subunit of RNAP. Wild-type MC4122 *Escherichia coli* cells were grown overnight in LB medium, subcultured into M9 medium (1% cultures), grown to early log phase (~0.05 absorbance at 600 nM) and transferred into wells for growth inhibition experiments. The bacterial growth in the absence or presence of synthetic L-peptides (250μM final concentration) was continuously monitored at 600 nM.

The assumption was that the peptides would bind to the σ-subunits, interfere with interactions of σ with β' essential for RNAP assembly, diminish RNAP holoenzyme formation, block gene transcription, and consequently inhibit bacterial growth. Indeed, peptide 4, which represents a σ-factor binding site on β' and exhibits the strongest binding ability to σ-subunits, completely stopped bacterial growth for more than 8 hours. Fig. 5. In good correspondence with the binding data, peptide 1 showed a significant inhibition of bacterial growth, whereas peptide 2 demonstrated only slight growth retardation potential. Peptide 3, which did not display a specific binding to the σ-subunits, did not interfere with bacterial growth at all. Fig. 5.

It should be noted that the peptide sequence representing σ-factor binding site on β'-subunit of RNAP is highly conserved among different bacterial species. Therefore, the prospective therapeutic agents, which could be designed based on described above principles, should have a broad anti-microbial specificity. The development of resistance to this type of anti-bacterial drugs should represent a very challenging task since it would require simultaneous mutations in two binding sites of at least two interacting RNAP subunits and both of these mutations would have to preserve the essential for RNAP assembly binding specificity.

Utilizing random peptide phage display libraries, we identified part of the conserved region A (amino acid residues 60-135) as a σ-binding region of β'-subunit. For precise identification (fine mapping) of σ-binding site on β' within β'-σ sub-assembly region, we developed a highly specific peptide- protein binding ELISA employing panel of synthetic biotinilated L-peptides derived from amino acid sequence of σ-binding region of β' as well as purified σ-proteins. Synthetic peptide 4 representing a 22-residue fragment within σ-binding region on β' (amino acid residues 69-90) exhibited a superior binding ability to the purified σ70 and σ38 subunits. Thus, β' sequence between Glu69 and Val90 represents a σ-binding site on β'-subunit. Furthermore, synthetic peptide 4 completely blocked bacterial growth for more than 8 hours, which was a duration time for *in vitro* growth inhibition experiments. Exposure to peptide 4 was apparently lethal for bacterial cells, since our attempt to recover living bacterial cells was not successful.

Identified peptide 4 sequence with higher binding ability toward the σ-subunit comprises the minimal subunit-subunit binding fragment. Peptide 4, representing the minimal subunit-subunit binding fragment is utilized as a peptide prototype for antibacterial drug design employing D-peptide ligand design strategy as in Example 2. A combination of the computational approach and combinatorial chemistry of small organic molecules are employed for translation of the structural information derived from the L-peptide sequences comprising minimal subunit-subunit binding fragments into small organic molecule antibacterial drug candidates.

The following steps are undertaken.

Homology modeling. This step consists of homology modeling of a 76 amino acid residue sigma-binding region acting as a mimic of β'-subunit of RNAP (SEQ. ID No. 4). The homology model is utlized to determine conformational information on the sigma-binding region and establish the 3D-positions of the amino acids essential for binding of the σ- subunit. Homology modeling is performed using technology from Insight II program (Molecular Simulations, Inc., San Diego, CA) and the Insight II-Homology module.

Building of an active site map. An active site map is derived from the homology model utilizing Ludi technology present in Cerius2 Structure-Based Focusing module software (Molecular Simulations, Inc., San Diego, CA). Structural-functional information on the peptide 4 (SEQ. ID No. 8) comprising the minimal σ-binding fragment of the β'-subunit is used to edit the active site map to dissect essential query features that will be translated into 3D pharmacophores. The preferable computational approach is a structure-based drug design utilizing complementary L-peptide/D-peptide pairs as a mimic of the receptor complex (see above).

High throughput screening using combinatorial libraries of small organic molecules. Peptide-protein binding ELISA is utilized for high throughput screening of commercially available random combinatorial libraries of small organic molecules. During this process the best small molecule competitors (Ki<50 nM) for binding of peptide 4 to the σ-subunit are selected and structurally identified. Five combinatorial libraries of small organic molecules that have established potential in generating hits from peptide-based screening systems are preferable for these experiments (see above). The hit discovery rates for these libraries during translation of 3D peptide-derived structural information into selection of small molecule drug candidates are within the range of 0.1-1.0%. Identification of at least 100 hits represents a sufficient number of hits identified by random screening in order to attempt to develop 3D pharmacophore models utilizing Catalyst technology

(Molecular Simulations, San Diego, CA). Subsequently, virtual screening of a database of commercially available compounds (ACD) against the Catalyst pharmacophore model is performed.

Virtual screening. This step would comprise virtual screening of a database of over 65,000 of commercially available compounds (ACD) against 3D pharmacophores derived in the previous step. HipHop software (Molecular Simulations, Inc., San Diego, CA) is utilized to perform a feature-based alignment of a collection of compounds onto a pharmacophore hypothesis. HipHop is used to match features, such as surface-accessible hydrophobes, charged or ionizable groups, or surface-accessible hydrogen bond donors or acceptors, against candidate molecules or searches of 3D databases. Ludi software (Molecular Simulations, Inc., San Diego, CA) is employed to fit molecules into the active site of a receptor by identifying and matching complementary polar and hydrophobic groups. An empirical scoring function is used to prioritize the hits. Ludi/ACD links the design tools of Ludi to MDL's Available Chemicals Directory and provides access to over 65,000 commercially available compounds with well defined structures. Approximately 100 small molecule drug candidates with the most promising binding ability should be identified (targeted sub-libraries of small molecule drug prototypes). The selected compounds will then be tested experimentally employing peptide-protein binding ELISA in order to confirm the 3D pharmacophore hypothesis and validate the selected structure-based design approach. The specified above quantitative criteria should be utilized for comparison of screening results of small molecule libraries and selection of the best drug candidate. The screening process is considered completed when at least 10 compounds with Ki<10nM in a peptide-protein binding ELISA have been identified and at least one small organic molecule library with a hit discovery rate 10% or more has been selected.

Additional considerations for targeting of the conserved region A on β'-subunit. A peptide fragment of conserved region A of β'-subunit (amino acid residues between Met29-Cys58) was identified as both DNA and RNA binding region of RNAP in a ternary elongation complex during RNA synthesis in the process of

gene transcription (Nudler et al., 1998). Protein-DNA cross-linking has implicated sequence of β' between Met29 and Met102 within conserved region A as participating in critical protein-DNA interactions during transcription (Nudler et al., 1996). Therefore, targeting this region of β'-subunit for antibacterial drug design is particularly attractive since, in addition to interference with subunit-subunit interactions, potential drug may interfere also with protein-nucleic acid interactions essential for function of a tertiary elongation complex, block RNA synthesis, abolish gene transcription and kill bacteria.

EXAMPLE 2

IDENTIFICATION OF D-PEPTIDE LIGANDS EMPLOYING MIRROR IMAGE PHAGE DISPLAY METHOD

This example of a proposed method of antibacterial drug design illustrates one possible approach that is targeted at blocking σ-β′ interaction during RNAP assembly. This approach also could be applied to interactions between other subunits of RNAP such as α, β, and β′. The synthetic D-peptide ligands developed in this example will bind to σ-contact sites on β′ subunits, interfere with the interactions of σ with β′ that are essential for RNAP assembly, diminish RNAP holoenzyme formation, block gene transcription, and consequently inhibit bacterial growth.

The combinatorial chemistry approach using either random or target-biased libraries of molecules for screening and selection of molecules with desired binding specificity is widely used for the purpose of structure-activity analysis and drug discovery. Among other genetically encoded libraries, random peptide bacteriophage display libraries may be used for this purpose. Scott and Smith (1990); Devlin, *et al.* (1990); Cwirla, *et al.* (1990). The method is based on repetitive synthesis and rescreening of peptides with desired binding specificity toward selected target molecules. The repetitive amplification of peptides interacting with target molecules in subsequent rounds of selection typically leads to the isolation and identification of specific peptide binding from a large random pool of peptide sequences displayed on phage surfaces. However, one of the major limitations of the

application of peptide phage display library screening for the purpose of drug discovery is that the resultant L-peptide ligands are subject to degradation by naturally occurring enzymes such as proteases and peptidases. Furthermore, peptides composed of naturally occurring L-amino acids (L-peptides) can induce a vigorous humoral immune response that would impair their biological activity. To overcome this drawback, a genetically encoded peptide phage display library is used for the identification of D-peptide ligands with binding specificity for the contact sites on subunits of RNAP.

The approach is based on the fact that the three-dimensional structure of D-proteins and D-peptides is the mirror image of the structure of the corresponding L-proteins and L-peptides. The D-enantiomeric peptide representing the selected peptide sequence of a natural L-protein target is prepared by chemical synthesis and used to isolate the L-peptide ligands that specifically interact with it from a peptide phage display library. Subsequently, the D-enantiomeric form of the isolated L-peptide ligands is prepared by chemical synthesis. The selection is performed in an achiral solvent, for example water, and the interaction between the L-peptide and the D-peptide does not require any chiral cofactors. Consequently, the D-enantiomers of the newly isolated L-peptide ligands specifically interact with and bind to the target protein of the natural, L-amino acid configuration. Thus, because of the mirror-image relation between the three-dimensional structures of peptide ligands for L-and D-enantiomeric proteins, the identification of phage-displayed L-peptides that bind to the D-enantiomer of a target protein molecule also provides the sequence of D-peptide ligands that bind to the natural L-protein target. Schumacher, *et al.* (1996).

Using this technique to identify the D-peptide ligands that bind to the sigma factor contact site on the β′ subunit of bacterial RNAP, the following steps are followed:

1) Chemical synthesis of the D-enantiomeric form of a σ- binding L-peptide, preferably the most efficient binding peptide.

2) Bioaffinity selection of L-peptide ligands for the D-enantiomer of the σ- binding L-peptide from the peptide phage display libraries. The affinity selection protocol is similar to the previously described protocol for biopanning and identification of σ^{38}-binding peptides.

3) Identification of the most potent L-peptide ligands for the D-enantiomer of the σ- binding L-peptide employing direct and competition ELISA assays with biotinilated peptides.

4) Chemical synthesis of the D-enantiomers forms of the most potent L-peptide ligands for the D-enantiomer of the σ- binding L-peptide.

5) Evaluation of the β'-binding activities of the D-enantiomeric forms of the L-peptide ligands for the D-enantiomer of the σ- binding L-peptide employing direct and competition ELISA assays using biotinilated synthetic D-peptide ligands and the β'-protein insert containing virions.

6) Identified synthetic D-peptide ligands with anticipated binding specificity toward the σ-contact site on β' are tested for their ability to inhibit the bacterial growth *in vitro* in an experimental protocol as described for σ-binding synthetic L-peptides. The comparison of the inhibitory potential of synthetic D-peptide ligands may be based on independently determined for each peptide ID_{50} doses. Initial testing is performed using *E. coli* cells, and subsequently, screening is expanded to include other bacterial strains.

EXAMPLE 3

TARGETING β- AND β'-BINDING SITES ON THE α-SUBUNIT

Peptide fragments of the conserved region A (amino acid residues 30 to 55) and conserved region B (amino acid residues 61 to 76) of the α-subunit are involved in direct interactions with the β-subunit during RNAP core assembly. Heyduck, *et al.* (1996) and Wang, *et al.*, (1997). Peptide fragments of both conserved region C (amino acid residues 175-185) and conserved region D (amino acid residues

195-210) of the α-subunit have been implicated in direct interactions with the β′-subunit during formation of the tertiary complex α2ββ′. Heyduck, *et al.* (1996).

The following six overlapping L-peptides derived from a protein sequence of the conserved regions A and B and conserved regions C and D of the a-subunit are chemically synthesized using methods known in the art:

Peptide 6: Pro Leu Glu Arg Gly Phe Gly His Thr Leu Gly Asn Ala Leu Arg Arg Ile Leu Leu Ser Met Pro Gly Cys (SEQ. ID No. 18) (PLERGFGHTLGNALRRILLSMPGC, aa 30-54)

Peptide 7: Cys Ala Val Thr Glu Val Glu Ile Asp Gly Val Leu His Glu Tyr Ser Thr Lys Glu Gly Val Gln Glu Asp Ile (SEQ. ID No. 19) (CAVTEVEIDGVLHEYSTKEGVQEDI, aa 54-78)

Peptide 8: Asn Ala Leu Arg Arg Ile Leu Leu Ser Met Pro Gly Cys AlaVal Thr Glu Val Glu Ile Asp Gly Val Leu (SEQ. ID No. 20) (NALRRILLSMPGCAVTEVEIDGVL, aa 41-65)

Peptide 9: Leu Val Asp Ala Cys Tyr Ser Pro Val Glu Arg Ile Ala Tyr Asn Val Glu Ala (SEQ. ID No. 21) (LVDACYSPVERIAYNVEA, aa 172-189)

Peptide 10: Ala Ala Arg Val Glu Gln Arg Thr Asp Leu Asp Lys Leu Val Ile Glu Met Glu Thr Asn Gly Thr (SEQ. ID No. 22) (ARVEQRTDLDKLVIEMETNGT, aa 190-210)

Peptide 11: Val Glu Arg Ile Ala Tyr Asn Val Glu Ala Ala Arg Val Glu Gln Arg Thr Asp Leu Asp Lys Leu Val (SEQ. ID No. 23) (VERIAYNVEAARVEQRTDLDKLV, aa 180-201)

In order to identify the minimal binding fragment, six chemically synthesized overlapping L-peptides are biotinilated and subjected to peptide-protein binding analysis employing experimental methodology described in the Example 1 for a fine mapping of the β'-σ contact site. The peptide-protein binding ELISA of the

peptides 6-8 would utilize the β-subunit as a target protein and therefore should yield a minimal binding site for β-subunit on α-subunit. The peptide-protein binding ELISA of the peptides 9-11 would utilize the β'-subunit as a target protein and therefore should yield a minimal binding site for β'-subunit on the α-subunit. Identified L-peptides with higher binding ability would comprise minimal subunit-subunit binding fragments. The minimal subunit-subunit binding fragments are utilized as a peptide prototype for antibacterial drug design employing D-peptide ligand design strategy as in Example 2 as well as combination of computational approach and combinatorial chemistry of small organic molecules for structure-guided drug design as in Example 1. The peptide fragment of the conserved regions A and B: Pro Leu Glu Arg Gly Phe Gly His Thr Leu Gly Asn Ala Leu Arg Arg Ile Leu Leu Ser Met Pro Gly Cys AlaVal Thr Glu Val Glu Ile Asp Gly Val Leu His Glu Tyr Ser Thr Lys Glu Gly Val Gln Glu Asp Ile (SEQ. ID No. 24)
(PLERGFGHTLGNALRRILLSMPGCAVTEVEIDGVLHEYSTKEGVQEDI, aa 30-78) and the peptide fragment of the conserved regions C and D: Leu Val Asp Ala Cys Tyr Ser Pro Val Glu Arg Ile Ala Tyr Asn Val Glu Ala Ala Arg Val Glu Gln Arg Thr Asp Leu Asp Lys Leu Val Ile Glu Met Glu Thr Asn Gly Thr (SEQ. ID No. 25)
(LVDACYSPVERIAYNVEAARVEQRTDLDKLVIEMETNGT, aa 175-210) are employed in the homology search step of computational approach as in Example 1.

EXAMPLE 4

TARGETING THE α- AND β′-BINDING SITES ON THE β-SUBUNIT

A portion of the conserved regions H and I (amino acid residues 907-1246) of the β-subunit, and particularly the N-terminal part of the conserved region I (amino acid residues 1115-1246) are involved in the binding of the β-subunit to the α-subunit during RNAP core assembly. Wang, *et al.* (1997). In addition, a portion of conserved region I (amino acid residues 1247-1342) of the β-subunit are involved in the recruitment of the β′-subunit to the α2β tertiary complex to form the tertiary complex α2ββ′. Wang, *et al.* (1997). Precise identification of the α-β and β-β` binding sites on the β-subunit will be performed employing peptide-protein binding

ELISA using biotinilated synthetic L-peptides derived from the primary sequences of corresponding binding regions and purified α and β′ subunits.

The following nineteen overlapping L-peptides derived from a protein sequence of the conserved regions H and I of the β-subunit are chemically synthesized using methods known in the art:

Peptide 12: Thr His Leu Gly Met Ala Ala Lys Gly Ile Gly Asp Lys Ile Asn Ala Met Leu Lys Gln Gln Gln Glu Val (SEQ. ID No. 26) (THLGMAAKGIGDKINAMLKQQQEV, aa 1115-1138)

Peptide 13: Ala Lys Leu Arg Glu Phe Ile Gln Arg Ala Tyr Asp Leu Gly Ala Asp Val Arg Gln Lys Val Asp Leu Ser (SEQ. ID No. 27) (AKLREFIQRAYDLGADVRQKVDLS, aa 1139-1162)

Peptide 14: Gly Asp Lys Ile Asn Ala Met Leu Lys Gln Gln Gln Glu Val Ala Lys Leu Arg Glu Phe Ile Gln Arg Ala (SEQ. ID No. 28) (GDKINAMLKQQQEVAKLREFIQRA, aa 1125-1148)

Peptide 15: Thr Phe Ser Asp Glu Glu Val Met Arg Leu Ala Glu Asn Leu Arg Lys Gly Met Pro Ieu Ala Thr Pro Val (SEQ. ID No. 29) (TFSDEEVMRLAENLRKGMPIATPV, aa 1163-1186)

Peptide 16: Phe Asp Gly Ala Lys Glu Ala Glu Ile Lys Glu Leu Leu Lys Leu Gly Asp Leu Pro Thr Ser Gly Gln Ile (SEQ. ID No. 30) (FDGAKEAEIKELLKLGDLPTSGQI, aa 1187-1210)

Peptide 17: Arg Leu Tyr Asp Gly Arg Thr Gly Glu Gln Phe Glu Arg Pro Val Thr Val Gly Tyr Met Tyr Met Leu Lys (SEQ. ID No. 31) (RLYDGRTGEQFERPVTVGYMYMLK, aa 1211-1234)

Peptide 18: Arg Pro Val Thr Val Gly Tyr Met Tyr Met Leu Lys Leu Asn His Leu Val Asp Asp Lys Met His Ala Arg (SEQ. ID No. 32) (RPVTVGYMYMLKLNHLVDDKMHAR, aa 1223-1246)

Peptide 19: Tyr Asp Leu Gly Ala Asp Val Arg Gln Lys Val Asp Leu Ser Thr Phe Ser Asp Glu Glu Val Met Arg Leu (SEQ. ID No. 33) (YDLGADVRQKVDLSTFSDEEVMRL, aa 1149-1172)

Peptide 20: Ala Glu Asn Leu Arg Lys Gly Met Pro Ieu Ala Thr Pro Val Phe Asp Gly Ala Lys Glu Ala Glu Ile Lys Glu Leu (SEQ. ID No. 34) (AENLRKGMPIATPVFDGAKEAEIKEL, aa 1173-1198)

Peptide 21: Leu Lys Leu Gly Asp Leu Pro Thr Ser Gly Gln Ile Arg Leu Tyr Asp Gly Arg Thr (SEQ. ID No. 35) (LKLGDLPTSGQIRLYDGRT, aa 1199-1217)

Peptide 22: Thr Gly Glu Gln Phe Glu Arg Pro Val Thr Val Gly Tyr Met Tyr Met Leu Lys Leu Asn His Leu (SEQ. ID No. 36) (GEQFERPVTVGYMYMLKLNHL, aa 1218-1238)

Peptide 23: : Ser Thr Gly Ser Tyr Ser Leu Val Thr Gln Gln Pro Leu Gly Gly Lys Ala Gln Phe Gly (SEQ. ID No. 37) (STGSYSLVTQQPLGGKAQFG, aa 1247-1266)

Peptide 24: Gly Gln Arg Phe Gly Glu Met Glu Val Trp Ala Leu Glu Ala Tyr Gly Ala Ala Tyr Thr Leu Gln Glu Met (SEQ. ID No. 38) (GQRFGEMEVWALEAYGAAYTLQEM, aa 1267-1290)

Peptide 25: Met Leu Thr Val Lys Ser Asp Asp Val Asn Gly Arg Thr Lys Met Tyr Lys Asn Ile Val Asp Gly (SEQ. ID No. 39) (LTVKSDDVNGRTKMYKNIVDG, aa 1291-1311)

Peptide 26: Asn His Gln Met Glu Pro Gly Met Pro Glu Ser Phe Asn Val Leu Leu Lys Glu Ile Arg Ser Leu Gly (SEQ. ID No. 40) (NHQMEPGMPESFNVLLKEIRSLG, aa 1312-1334)

Peptide 27: Met Pro Glu Ser Phe Asn Val Leu Leu Lys Glu Ile Arg Ser Leu Gly Ile Asn Ile Glu Leu Glu Asp Glu (SEQ. ID No. 41) (MPESFNVLLKEIRSLGINIELEDE, aa 1319-1342)

Peptide 28: Thr Gln Gln Pro Leu Gly Gly Lys Ala Gln Phe Gly Gly Gln Arg Phe Gly Glu Met Glu Val (SEQ. ID No. 42)
(TQQPLGGKAQFGGQRFGEMEV, aa 1255-1275)

Peptide 29: Val Trp Ala Leu Glu Ala Tyr Gly Ala Ala Tyr Thr Leu Gln Glu Met Leu Thr Val Lys Ser Asp Asp Val (SEQ. ID No. 43)
(VWALEAYGAAYTLQEMLTVKSDDV, aa 1275-1298)

Peptide 30: Val Asn Gly Arg Thr Lys Met Tyr Lys Asn Ile Val Asp Gly Asn His Gln Met Glu Pro Gly (SEQ. ID No. 44)
(VNGRTKMYKNIVDGNHQMEPG, aa 1298-1318)

In order to identify the minimal binding fragment, nineteen chemically synthesized overlapping L-peptides are biotinilated and subjected to peptide- protein binding analysis employing experimental methodology described in the Example 1 for a fine mapping of the β'-σ contact site. The peptide-protein binding ELISA of the peptides 12-22 would utilize the α-subunit as a target protein and therefore should yield a minimal binding site for the α-subunit on β-subunit. The peptide-protein binding ELISA of the peptides 23-30 would utilize the β'-subunit as a target protein and therefore should yield a minimal binding site for β'-subunit on β-subunit. Identified L-peptides with higher binding ability would comprise minimal subunit-subunit binding fragments. The minimal subunit-subunit binding fragments are utilized as a peptide prototype for antibacterial drug design employing D-peptide ligand design strategy as in Example 2 as well as combination of computational approach and combinatorial chemistry of small organic molecules for structure-guided drug design as in Example 1. Two peptide fragments of the conserved regions H and I of the β-subunit are employed in the Homology Search step of computational approach as in Example 1. Peptide fragment 1: Thr His Leu Gly Met Ala Ala Lys Gly Ile Gly Asp Lys Ile Asn Ala Met Leu Lys Gln Gln Gln Glu Val Ala Lys Leu Arg Glu Phe Ile Gln Arg Ala Tyr Asp Leu Gly Ala Asp Val Arg Gln Lys Val Asp Leu Ser Thr Phe Ser Asp Glu Glu Val Met Arg Leu Ala Glu Asn Leu Arg Lys Gly Met Pro Ieu Ala Thr Pro Val Phe Asp Gly Ala Lys Glu Ala Glu Ile Lys Glu Leu Leu Lys Leu Gly

Asp Leu Pro Thr Ser Gly Gln Ile Arg Leu Tyr Asp Gly Arg Thr Gly Glu Gln Phe Glu Arg Pro Val Thr Val Gly Tyr Met Tyr Met Leu Lys Leu Asn His Leu Val Asp Asp Lys Met His Ala Arg (SEQ. ID No. 45)

(THLGMAAKGIGDKINAMLKQQQEVAKLREFIQRAYDLGADVRQKVDLSTFS DEEVMRLAENLRKGMPIATPVFDGAKEAEIKELLKLGDLPTSGQIRLYDGRTG EQFERPVTVGYMYMLKLNHLVDDKMHAR, aa 1115-1246); Peptide fragment 2: Ser Thr Gly Ser Tyr Ser Leu Val Thr Gln Gln Pro Leu Gly Gly Lys Ala Gln Phe Gly Gly Gln Arg Phe Gly Glu Met Glu Val Trp Ala Leu Glu Ala Tyr Gly Ala Ala Tyr Thr Leu Gln Glu Met Leu Thr Val Lys Ser Asp Asp Val Asn Gly Arg Thr Lys Met Tyr Lys Asn Ile Val Asp Gly Asn His Gln Met Glu Pro Gly Met Pro Glu Ser Phe Asn Val Leu Leu Lys Glu Ile Arg Ser Leu Gly Ile Asn Ile Glu Leu Glu Asp Glu (SEQ. ID No. 46)

(STGSYSLVTQQPLGGKAQFGGQRFGEMEVWALEAYGAAYTLQEMLTVKSD DVNGRTKMYKNIVDGNHQMEPGMPESFNVLLKEIRSLGINIELEDE, aa 1247-1342).

Additional considerations for targeting conserved region I of β-subunit.

Conserved region I of β-subunit (amino acid residues between Met1232-Met1304) was identified as both DNA and RNA binding region of RNAP in a ternary elongation complex during RNA synthesis in the process of gene transcription (Nudler et al., 1998). Protein-DNA cross-linking has implicated a sequence between Met1230 and Met1273 within conserved region I as participating in critical protein-DNA interactions during transcription (Nudler et al., 1996, 1998). Therefore, targeting this region of β-subunit for antibacterial drug design is particularly attractive since, in addition to interference with subunit-subunit interactions, potential drug may interfere also with protein-nucleic acid interactions essential for function of a ternary elongation complex, block RNA synthesis, and abolish gene transcription.

EXAMPLE 5

TARGETING RNAP CORE BINDING SITE ON THE σ-SUBUNIT

Targeting a core-binding site on the σ-subunit for antibacterial drug design should offer several potential benefits. Eukaryotic cells do not employ σ-subunits for initiation of gene transcription. Therefore, the application of σ-targeted antibacterial drugs should not lead to host toxicity. As an enzyme that transcribes DNA processively, core RNAP binds tightly to single-stranded or double-stranded DNA. Strauss, *et al.* (1980). Binding of the σ-subunit to core RNAP decreases its affinity for nonspecific DNA binding. deHaseth, *et al.* (1978) and Lhoman, *et al.* (1980). It also increases its affinity for specific promoter DNA binding. Hinkle & Chamberlin (1972) and Chamberlin (1976). However, although the σ70 protein contains two specific DNA-binding domains that recognize the two conserved regions of prokaryotic promoters, intact σ70 protein does not bind to DNA. Thus, the DNA-binding domains of intact σ70 are buried within other regions of the protein. Association of σ70 with core RNAP induces conformational changes in σ70 that unmask the DNA-binding domains of the σ-subunit. Dombroski, *et al.* (1992). Similarly, interactions of other regulatory DNA-binding proteins with small molecule effectors were required to expose their DNA-binding domains: CAP requires cAMP and *trpR* repressor requires tryptophan for sequence-specific DNA binding. Saxe & Revzin (1979) and Gunsalus & Yanofsky (1980). It is conceivable that binding of small molecule drugs to the core-binding site of σ70 would induce conformational changes in σ70 the unmask the DNA-binding domains of the σ-subunit. Activated σ70 should bind to the specific promoter DNA sequences, prevent promoter recognition and binding to the promoter of RNAP holoenzyme molecules thus blocking transcription initiation. Therefore, application of antibacterial drugs targeting core-binding sites on σ-subunit should prevent assembly of the new RNAP holoenzyme molecules and block function of the existing RNAP holoenzyme molecules since the σ70/small molecule complex may act as a gene transcription repressor as well.

A fragment of conserved region 2.1 (amino acid residues 361-390) of σ70 subunit was identified as a RNAP core binding site of σ-subunit. Lesley & Burgess (1989) and Lonetto, *et al.* (1992). Subsequently, mutational analysis demonstrated that σ70 mutants with the amino acid sequence 361-374 deleted still exhibited sufficient core binding ability to produce transcriptionally competent RNAP holoenzyme. Kumar, *et al.* (1995). Peptide 4 from Example 1, above, representing a σ-binding site on the β′-subunit, and the σ70-derived peptide 375-390, comprising a core binding sequence of the σ70-subunit, may bind to each other providing a receptor/ligand mimic of the corresponding subunit-subunit contact site. Cross-competition ELISA experiments using corresponding synthetic peptides and purified RNAP subunits are performed to test this hypothesis.

The following three overlapping L-peptides derived from the peptide sequence 361-390 of σ70 (SEQ. ID No. 51) are chemically synthesized using methods known in the art:

Peptide 31: Ile Asn Arg Arg Met Ser Ile Gly Glu Ala Lys Ala Arg Arg Ala (SEQ. ID No. 48) (INRRMSIGEAKARRA, aa 361-375)

Peptide 32: Ala Lys Lys Glu Met Val Glu Ala Asn Leu Arg Leu Val Ile Ser Ile (SEQ. ID No. 49) (AKKEMVEANLRLVISI, aa 375-390)

Peptide 33: Met Ser Ile Gly Glu Ala Lys Ala Arg Arg Ala Lys Lys Glu Met Val Glu Ala (SEQ. ID No. 50) (IGEAKARRAKKEMVEA, aa 367-382)

In order to identify the minimal core-binding fragment within the peptide sequence 361-390 of σ70, three chemically synthesized overlapping L-peptides are biotinilated and subjected to peptide-protein binding analysis employing experimental methodology described in the example 1 for a fine mapping of the β'-σ contact site. The peptide-protein binding ELISA of the peptides 31-33 would utilize the β'-subunit as a target protein and therefore should yield a minimal binding site for β'-subunit on σ-subunit. Identified L-peptide with higher binding ability would comprise the minimal subunit-subunit binding fragment. The minimal subunit-

subunit binding fragments are utilized as a peptide prototype for antibacterial drug design employing D-peptide ligand design strategy as in Example 2, as well as combination of computational approach and combinatorial chemistry of small organic molecules for structure-guided drug design as in Example 1.

The peptide sequence of σ70 (Ile Asn Arg Arg Met Ser Ile Gly Glu Ala Lys Ala Arg Arg Ala Lys Lys Glu Met Val Glu Ala Asn Leu Arg Leu Val Ile Ser Ile; (SEQ. ID No. 51) INRRMSIGEAKARRAKKEMVEANLRLVISI, aa 361-390) is employed in the homology search step of the computational approach as in Example 1.

EXAMPLE 6

TARGETING NUCLEIC ACID-BINDING SITES ON THE β'-SUBUNIT

Two nucleic acid-binding sites, one within conserved region A (amino acid residues between Met1 and Met 102) and another within conserved region C (amino acid residues between Met298 and Met230), were identified on the β'-submit of RNAP (Nudler et al., 1996, 1998). More specifically nucleic acid-binding sites were mapped in the two areas within the sequence between Met1 and Met102: between amino acids Met1-Met29 and amino acid Met29-Cys58 (Nudler et al., 1998). Synthetic L-peptides derived from amino acid sequences of the corresponding nucleic acid-binding sites will be tested for DNA-binding ability and the minimal DNA-binding peptide fragment within each nucleic acid-binding sequence will be determined. Identified minimal DNA-binding peptide fragments will be utilized as a receptor prototype in order to generate specific D-peptide ligands employing a mirror image phage display method. Corresponding synthetic L-peptide/D-peptide pairs will be utilized as a receptor/ligand mimic for a structure-guided drug design employing a computational approach as well as combinatorial small molecule libraries as described in Example 1.

The thirteen overlapping peptides derived from putative nucleic acid-binding sequences of the conserved region A (SEQ. ID No. 62) (aa 1-61) and

conserved region C (SEQ. ID No. 63) (aa 298-330) of the β'-subunit are synthesized chemically by methods known in the art.

Peptide 34: Met Lys Asp Leu Leu Lys Phe Leu Lys Ala Gln Thr Lys Thr Glu Glu Phe Asp Ala Ile Lys Ile Ala (SEQ. ID No. 54) (MKDLLKFLKAQTKTEEFDAIKIA, aa 1-23)

Peptide 35: Ile Ala Leu Ala Ser Pro Asp Met Ile Arg Ser Trp Ser Phe Gly Glu Val Lys Lys Pro Glu Thr Ile (SEQ. ID No. 55) (ALASPDMIRSWSFGEVKKPETI, aa 23-44)

Peptide 36: Thr Ile Asn Tyr Arg Thr Phe Lys Pro Glu Arg Asp Gly Leu Phe Cys Ala Arg Ile (SEQ. ID No. 56) (INYRTFKPERDGLFCARI, aa 44-61)

Peptide 37: Val Lys Lys Pro Glu Thr Ile Asn Tyr Arg Thr Phe Lys Pro Glu Arg Asp Gly Leu Phe Cys (SEQ. ID No. 57) (VKKPETINYRTFKPERDGLFC, aa 38-58)

Peptide 38: Thr Glu Glu Phe Asp Ala Ile Lys Ile Ala Leu Ala Ser Pro Asp Met Ile Arg Ser Trp Ser Phe Gly Glu Val (SEQ. ID No. 58) (TEEFDAIKIALASPDMIRSWSFGEV, aa 14-38)

Peptide 39: Met Leu Gln Glu Ala Val Asp Ala Leu Leu Asp Asn Gly Arg Arg Gly Arg Ala (SEQ. ID No. 59) (MLQEAVDALLDNGRRGRA, aa 298-315)

Peptide 40: Ala Ile Thr Gly Ser Asn Lys Lys Pro Leu Lys Ser Leu Ala Asp Met (SEQ. ID No. 60) (AITGSNKRPLKSLADM, aa 315-330)

Peptide 41: Leu Asp Asn Gly Arg Arg Gly Arg Ala Ile Thr Gly Ser Asn Lys Lys Pro Leu Lys Ser Leu (SEQ. ID No. 61) (LDNGRRGRAITGSNKRPLKSL, aa 307-327)

The amino acid sequence between R60 and Met102 is covered by synthetic peptides 1-5 in Example 1 that have been designed for a fine mapping of the s-binding site on β'-subunit. In order to identify the minimal nucleic acid-binding

fragment on the β'-subunit, thirteen chemically synthesized overlapping L-peptides are biotinilated and subjected to peptide-DNA binding analysis employing gel-shift assay and/or ELISA technique. The binding analysis of the peptides uses the Escherichia coli DNA as a target and therefore should yield a minimal binding site for DNA on β'-subunit. Identified L-peptide with higher binding ability would comprise the minimal DNA-binding fragment. The minimal DNA-binding fragments are utilized as a peptide prototype for antibacterial drug design employing D-peptide ligand design strategy as in Example 2, as well as combination of computational approach and combinatorial chemistry of small organic molecules for structure-guided drug design as in Example 1.

The putative nucleic acid-binding sequences of the conserved region A: Met LysAsp Leu Leu Lys Phe Leu Lys Ala Gln Thr Lys Thr Glu Glu Phe Asp Ala Ile Lys Ile Ala Leu Ala Ser Pro Asp Met Ile Arg Ser Trp Ser Phe Gly Glu Val Lys Lys Pro Glu Thr Ile Asn Tyr Arg Thr Phe Lys Pro Glu Arg Asp Gly Leu Phe Cys Ala Arg Ile (SEQ. ID No. 62)
(MKDLLKFLKAQTKTEEFDAIKIALASPDMIRSWSFGEVKKPETINYRTFKPER DGLFCARI, aa 1-61) and conserved region C: Met Leu Gln Glu Ala Val Asp Ala Leu Leu Asp Asn Gly Arg Arg Gly Arg Ala Ile Thr Gly Ser Asn Lys Lys Pro Leu Lys Ser Leu Ala AspMet (SEQ. ID No. 63)
(MLQEAVDALLDNGRRGRAITGSNKRPLKSLADM, aa 298-330) of the β'-subunit are employed in homology search step of the computational approach.

EXAMPLE 7

TARGETING NUCLEIC ACID-BINDING SITES ON THE β-SUBUNIT

Two amino acid fragments of the β-subunit were implicated as nucleic acid-binding sites of RNAP during transcription: the amino acid sequence between Met130 and Met239 of the conserved region B and the amino acid sequence between Met1230 and Met1304 of the conserved region I (Nudler et al., 1996, 1998). Synthetic L-peptides derived from amino acid sequences of the corresponding nucleic acid-binding sites will be tested for DNA-binding ability and the minimal DNA-

binding peptide fragment within each nucleic acid-binding sequence will be determined. Identified minimal DNA-binding peptide fragments will be utilized as a receptor prototype in order to generate specific D-peptide ligands employing a mirror image phage display method. Corresponding synthetic L-peptide/D-peptide pairs will be utilized as a receptor/ligand mimic for a structure-guided drug design employing computational approach as well as combinatorial small molecule libraries as in Example 1.

The following fifteen overlapping peptides derived from putative nucleic acid-binding sequences of the conserved region B (SEQ. ID No. 81) (aa130-239) and conserved region I (SEQ. ID No. 82) (aa 1230-1304) of the β'-subunit are synthesized chemically by methods known in the art:

Peptide 42: Met Thr Asp Asn Gly Thr Phe Val Ile Asn Gly Thr Glu Arg Val Ile Val Ser Gln Leu His Arg (SEQ. ID No. 66)
(MTDNGTFVINGTERVIVSQLHR, aa 130-151)

Peptide 43: Ser Pro Gly Val Phe Phe Asp Ser Asp Lys Gly Lys Thr His Ser Ser Gly Lys Val Leu Tyr Asn (SEQ. ID No. 67)
(SPGVFFDSDKGKTHSSGKVLYN, aa 152-173)

Peptide 44: Ala Arg Ile Ile Pro Tyr Arg Gly Ser Trp Leu Asp Phe Glu Phe Asp Pro Lys Asp Asn Leu Phe (SEQ. ID No. 68)
(ARIIPYRGSWLDFEFDPKDNLF, aa 174-195)

Peptide 45: Val Arg Ile Asp Arg Arg Arg Lys Leu Pro Ala Thr Ile Ile Leu Arg Ala Leu Asn Tyr Thr Thr (SEQ. ID No. 69)
(VRIDRRRKLPATIILRALNYTT, aa 196-217)

Peptide 46: Glu Gln Ile Leu Asp Leu Phe Phe Glu Lys Val Ile Phe Glu Ile Arg Asp Asn Lys Leu Gln Met (SEQ. ID No. 70)
(EQILDLFFEKVIFEIRDNKLQM, aa 218-239)

Peptide 47: Ser Trp Leu Asp Phe Glu Phe Asp Pro Lys Asp Asn Leu Phe Val Arg Ile Asp Arg Arg Arg Lys Leu Pro (SEQ. ID No. 71) (SWLDFEFDPKDNLFVRIDRRRKLP, aa 182-205)

Peptide 48: Val Ser Gln Leu His Arg Ser Pro Gly Val Phe Phe Asp Ser Asp Lys Gly Lys Thr His Ser (SEQ. ID No. 72) (VSQLHRSPGVFFDSDKGKTHS, aa 146-166)

Peptide 49: Ser Gly Lys Val Leu Tyr Asn Ala Arg Ile Ile Pro Tyr Arg Gly Ser Trp Leu (SEQ. ID No. 73) (SGKVLYNARIIPYRGSWL, aa 167-184)

Peptide 50: Leu Asp Phe Glu Phe Asp Pro Lys Asp Asn Leu Phe Val Arg Ile Asp Arg Arg Arg Lys Leu Pro (SEQ. ID No. 74) (LDFEFDPKDNLFVRIDRRRKLP, aa 184-205)

Peptide 51: Ala Thr Ile Ile Leu Arg Ala Leu Asn Tyr Thr Thr Glu Gln Ile Leu Asp Leu Phe Phe Glu Lys Val (SEQ. ID No. 75) (ATIILRALNYTTEQILDLFFEKV, aa 206-228)

Peptide 52: Met Tyr Met Leu Lys Leu Asn His Leu Val Asp Asp Lys Met His Ala Arg Ser Thr Gly Ser Tyr Ser Leu Val (SEQ. ID No. 76) (MYMLKLNHLVDDKMHARSTGSYSLV, aa 1230-1254)

Peptide 53: Thr Gln Gln Pro Leu Gly Gly Lys Ala Gln Phe Gly Gly Gln Arg Phe Gly Glu Met Glu Val Trp Ala Leu Glu (SEQ. ID No. 77) (TQQPLGGKAQFGGQRFGEMEVWALE, aa 1255-1279)

Peptide 54: Ala Tyr Gly Ala Ala Tyr Thr Leu Gln Glu Met Leu Thr Val Lys Ser Asp Asp Val Asn Gly Arg Thr Lys Met (SEQ. ID No. 78) (AYGAAYTLQEMLTVKSDDVNGRTKM, aa 1280-1304)

Peptide 55: Met His Ala Arg Ser Thr Gly Ser Tyr Ser Leu Val Thr Gln Gln Pro Leu Gly Gly Lys Ala Gln Phe Gly (SEQ. ID No. 79) (MHARSTGSYSLVTQQPLGGKAQFG, aa 1243-1266)

Peptide 56: Gly Gln Arg Phe Gly Glu Met Glu Val Trp Ala Leu Glu Ala Tyr Gly Ala Ala Tyr Thr Leu Gln Glu Met Leu (SEQ. ID No. 80) (GQRFGEMEVWALEAYGAAYTLQEML, aa 1267-1291)

In order to identify the minimal nucleic acid-binding fragment on the β-subunit, fifteen chemically synthesized overlapping L-peptides are biotinilated and subjected to peptide-DNA binding analysis employing gel-shift assay and/or ELISA technique. The binding analysis of the peptides would utilize the Escherichia coli DNA as a target and therefore should yield a minimal binding site for DNA on β-subunit. Identified L-peptide with higher binding ability would comprise the minimal DNA-binding fragment. The minimal DNA-binding fragments are utilized as a peptide prototype for antibacterial drug design employing D-peptide ligand design strategy as in Example 2 as well as combination of computational approach and combinatorial chemistry of small organic molecules for structure-guided drug design as in Example 1.

The following putative nucleic acid-binding sequences of the conserved region B: Met Thr Asp Asn Gly Thr Phe Val Ile Asn Gly Thr Glu Arg Val Ile Val Ser Gln Leu His Arg Ser Pro Gly Val Phe Phe Asp Ser Asp Lys Gly Lys Thr His Ser Ser Gly Lys Val Leu Tyr Asn Ala Arg Ile Ile Pro Tyr Arg Gly Ser Trp Leu Asp Phe Glu Phe Asp Pro Lys Asp Asn Leu Phe Val Arg Ile Asp Arg Arg Arg Lys Leu Pro Ala Thr Ile Ile Leu Arg Ala Leu Asn Tyr Thr Thr Glu Gln Ile Leu Asp Leu Phe Phe Glu Lys Val Ile Phe Glu Ile Arg Asp Asn Lys Leu Gln Met (SEQ. ID No. 81) (MTDNGTFVINGTERVIVSQLHRSPGVFFDSDKGKTHSSGKVLYNARIIPYRGS WLDFEFDPKDNLFVRIDRRRKLPATIILRALNYTTEQILDLFFEKVIFEIRDNKL QM, aa130-239) and conserved region I: Met Tyr Met Leu Lys Leu Asn His Leu Val Asp Asp Lys Met His Ala Arg Ser Thr Gly Ser Tyr Ser Leu Val Thr Gln Gln Pro Leu Gly Gly Lys Ala Gln Phe Gly Gly Gln Arg Phe Gly Glu Met Glu Val Trp Ala Leu Glu Ala Tyr Gly Ala Ala Tyr Thr Leu Gln Glu Met Leu Thr Val Lys Ser Asp Asp Val Asn Gly Arg Thr Lys Met (SEQ. ID No. 82) (MYMLKLNHLVDDKMHARSTGSYSLVTQQPLGGKAQFGGQRFGEMEVWAL

EAYGAAYTLQEMLTVKSDDVNGRTKM, aa 1230-1304) of the β'-subunit are employed in homology search step of the computational approach.

Although the invention has been described with reference to the examples above, it should be understood that various modifications can be made without departing from the spirit of the invention. Accordingly, the invention is limited only by the following claims. All publications, patents, and patent applications cited herein are hereby incorporated by reference.

REFERENCES

Baldino, C.M., et al., "Convergent Parallel Synthesis," Syn. Lett., (May 1997) pp. 488-490.

Bunin, B.A., et al., "The combinatorial synthesis and chemical and biological evaluation of a 1,4-benzodiazepine library," Proc. Natl. Acad. Sci. USA, (1994) **91**, p. 4708.

Bunin, B.A., et al., " " J. Am. Chem. Soc., (1992) **114**, pp. 10997.

Chamberlin, M. J., "Interaction of RNA Polymerase with DNA Template," RNA Polymerase, R. Losick and M. Chamberlin Eds., (Cold Spring Harbor, New York: Cold Spring Harbor Laboratory), (1976), pp. 159-191.

Coggins, J.R., et al., " ," Biochemistry, (1997) **16**, pp. 1111-1116.

Cull, M.G., et al., "Screening for receptor ligands using large libraries of peptides linked to the C terminus of the lac repressor," Proc. Natl. Acad. Sci. USA, (1992) **89**, p. 1865.

Cwirla, S.E., et al. "Peptides on phage: a vast library of peptides for identifying ligands," Proc. Natl. Acad. Sci. USA, (1990) **87**, pp. 6378-6381.

DeHaseth, P.L., et al., "Non-Specific Interactions of *Escherichia coli*, RNA Polymerase with Native and Denatured DNA: Differences in the Binding Behavior of Core and Holoenzyme," Biochemistry, (1978) **17**, pp. 1612-1622.

Devlin, J.J., et al., "Random peptide libraries: a source of specific protein binding molecules," Science, (1990), **249**, pp. 404-407.

Dombroski, A.J., et al., "Polypeptides Containing Highly Conserved Regions of Transcription Initiation Factor σ^{70} Exhibit Specificity of Binding to Promoter DNA," Cell, (1992) **70**, pp. 501-512.

Dooley, C.T., *et al.*, "An all D-amino acid opioid peptide with central analgesic activity from a combinatorial library," Science, (1994) **266**, p. 2019.

Evans, B.E., et al., "Methods for drug discovery: development of potent, selective, orally effective cholecystokinin antagonists," J. Med. Chem., (1988) **31**, p. 2235.

Fukuda, R., et al., "Subunits of RNA polymerase in function and structure; Maturation in vitro of core enzyme from Escherichia coli," J. Mol. Biol., (1974) **87**, pp. 523-540.

Glass, R.E., et al., "Genetic studies on the beta subunit of Escherichia coli RNA polymerase. IX. The role of the carboxy-terminus in enzyme assembly," Mol. Gen. Genet., (1986) **203**, pp. 492-495.

Gordon, E.M., et al., "Applications of combinatorial technologies to drug discovery. 2. Combinatorial organic synthesis, library screening strategies, and future directions," J. Med. Chem., (1994) **37**, p. 1385.

Greiner, D.P., "Binding of the σ^{70} protein to the core subunits of Escherichia coli RNA polymerase, studied by iron-EDTA protein footprinting," Proc. Natl. Acad. Sci. USA, (1996) **93**, pp. 71-75.

Gunsalus, R.P., et al., "Nucleotide Sequence and Expression of *Escherichia coli trpR*, The Structural Gene for the trp Aporepressor," Proc. Natl. Acad. Sci. USA, ( ) **77**, pp. 7117-7121.

Hancock, R.E.W. "Antibacterial peptides and the outer membranes of gram-negative bacilli," J. Med. Microbiol., (1997) **46**, pp. 1-3.

Hass, S.J. and Smith, G.P. Rapid sequencing of viral DNA from filamentous bacteriophage. Biotechniques, (1993) **15**(3), pp. 422-424, 426-428, 431.

Helmann, J.D., et al., "Structure and function of bacterial sigma factors," Annu. Rev. Biochem., (1988) **57**, pp. 839-872.

Heyduk, T.,et al, "Rapid epitope mapping by hydroxyl-radical protein footprinting: Determinants of RNA polymerase alpha subunit for interaction with beta, beta', and sigma subunits," Proc. Natl. Acad. Sci. USA, (1996) **93**, pp. 10162-10166.

Hillel, Z., et al., "Subunit topography of RNA polymerase from Escherichia coli. A cross-linking study with bifunctional reagents," Biochemistry, (1977) **16**, pp. 3334-3342.

Hinkle, D.C., et al., "Studies of the Binding of *Escherichia coli*, RNA Polymerase to DNA. 1. The Role of Sigma Subunit in Site Selection," J. Mol. Biol., (1972), **70**, pp. 157-185.

Hobd De Witt, S., et al. "'Diversomers': an approach to nonpeptide, nonoligomeric chemical diversity," Proc. Natl. Acad. Sci. USA, (1993), **90**, p. 6909.

Hogan, J.C., "Combinatorial Chemistry in Drug Discovery," Nature Biotechnology, (1997), **15**, pp. 328-330.

Hogan, J.C., "Directed Combinatorial Chemistry," Nature, (1996), **384**, pp. 17-19.

Houghton, R.A., " ," Proc. Natl. Acad. Sci USA, (1985) **82**, p. 5131.

Houghton, R.A., et al., " ," J. Pept. Prot. Res., (1986), **27**, p. 673.

Houghton, R.A., et al., " ," Nature, (1991) **354**, p. 64.

Ishihama, A., "Subunit assembly of Escherichia coli RNA polymerase," Advan. Biophys. (1981) **14**, pp. 1-35.

Ishihama, A., "Promoter selectivity of prokaryotic RNA polymerase," Trends Genet., (1988) **4**, pp. 282-286.

Ishihama, A., "Molecular assembly and functional modulation of Escherichia coli RNA polymerase," Adv. Biophys., (1990) **26**, pp. 19-31.

Ishihama, A., " Protein-protein communication within transcription apparatus," J. Bacteriol., (1993) **175**, pp. 2483-2489.

Kumar, A., et al., "A Partially Functional 245-Amino Acid Internal deletion Derivative of *Escherichia coli* σ^{70}," J. Bacteriol., (1995) **177**, pp. 5193-5196.

Lange, R.N., et al., "Identification of a central regulator of stationary-phase gene expression in Escherichia coli," Mol. Microbiol., (1991) **5**, 49-59.

Lesley, S.A., et al., "Characterization of the *Escherichia coli* Transcription Factor σ^{70}: Localization of a Region Involved in the Interaction with Core RNA Polymerase," Biochemistry, (1989) **28**, pp. 7728-7734.

Lesley, S.A., et al., "Use of in vitro protein synthesis from polymerase chain reaction-generated templates to study interaction of Escherichia coli transcription factors with core RNA polymerase and for epitope mapping of monoclonal antibodies," J. Biol. Chem., (1991) **266**, pp. 2632-2638.

Lohman, T.M., et al., "Use of Difference Boundary Sedimentation Velocity to Investigate Nonspecific Protein-nucleic Acid Interactions," Biochemistry, (1998) **19**, pp. 3516-3522.

Lonetto, M., et al., "The σ^{70} family: sequence conservation and evolutionary relationships," J. Bacteriol., (1992) **174**, 3843-3849.

Luo, J., et al., "Molecular Anatomy of the β′ subunit of the E. coli RNA polymerase: identification of regions involved in polymerase assembly," Genes Cells, (1996) **1**, pp. 819-827.

Mattheakis, L.C., et al., "An in vitro polysome display system for identifying ligands from very large peptide libraries," Proc.Natl.Acad.Sci.USA, (1994) **91**, p. 9022.

McMahan, S.A., et al., "Use of aryl azide cross-linkers to investigate protein-protein interactions: an optimization of important conditions as applied to Escherichia

coli RNA polymerase and localization of a sigma 70-alpha cross-link to the C-terminal region of alpha," Biochemistry, (1994) **33**, pp. 12092-12099.

Miyake, R., et al., "Mapping of the σ^{70} subunit contact sites on Escherichia coli-RNA polymerase with a σ^{70} conjungated chemical protease," Proc. Natl. Acad. Sci. USA, (1998) **95**, pp. 6021-6026.

Mulvey, M.R., et al., "Nucleotide sequence of katF of Escherichia coli suggests KatF protein is a novel σ transcription factor," Nucleic Acids Res., (1989) **17**, pp. 9979-9991.

Neu, H.C., "Infection problems for the 1990's - do we have an answer?" Scan.J.Infect.Dis.Suppl., (1993) **91**, pp. 7-13.

Nguyen, L.H., et al., *In vitro* functional characterization of overproduced Escherichia coli katF/rpoS gene product," Biochemistry, (1993) **32**, pp. 11112-11117.

Nudler, E., "Spatial Organization of Transcription Elongation Complex in *Escherichia coli*," Science, (1998) **281**, pp. 424-428.

Nudler, E., Avetissova, et al., "Transcription processivity; protein-DNA interactions holding together the elongation complex," Science, (1996) **273**, pp. 211-217.

Patek, M., et al., " ," Tetrahedron Lett., (1995), **36**, p. 22-27.

Pearson, W.R., et al., "Improved tools for biological sequence comparison," Proc.Natl.Acad.Sci.USA, (1988) **85**, pp. 2444-2448.

Peisach, E., et al., "Interaction of a Peptidomimetic Aminimide Inhibitor with Elastase," Science, (1995) **269**, pp. 66-69.

Peletskaya, E.N., et al., "Identification of peptide sequences that bind the Thomsen-Friedenreich cancer-associated glycoantigen from bacteriophage peptide display libraries," Mol. Diversity, (1996) **2**, 13-18.

Peletskaya, E.N..et al., "Characterization of peptides that bind the tumor-associated Thomsen-Friedenreich antigen selected from bacteriophage display libraries," J. Mol. Biol., (1997) **270**, pp. 373-384.

Puhler *et al.*, "Archaebacterial DNA-dependent RNA polymerases testify to the evolution of the eukaryotic nuclear genome," Proc. Natl. Acad. Sci. USA, (1989) **86**, p. 4569.

Saxe S., et al., "Cooperative Binding to DNA of Catabolite Activator Protein of *Escherichia coli*," Biochemistry, (1979) **18**, pp. 255-263.

Schumacher, T.N.M., et al. "Identification of D-peptide ligands through mirror-image phage display," Science (1996) **271**, pp. 1854-1957.

Scott, J.K.,et al., "Searching for peptide ligands with an epitope library," Science (1990) **249**, pp. 386-390.

Siegel, D.A., et al., " ," J. Mol. Biol., (1989) **206**, pp. 591-603.

Silver, L., et al., "Screening of natural products for antimicrobial agents," Eur.J.Clin.Microbiol.Infect.Dis., (1990) **9**, pp. 455-461.

Smith, G.P., et al., "Libraries of peptides and proteins displayed on filamentous phage," Methods Enzymol., (1993) **217**, pp. 228-257.

Strauss, H.S., et al., "Binding of *Escherichia coli* Ribonucleic acid polymerase Holoenzyme to a Bacteriophage T7 Promoter-containing Fragment: Evaluation of Promoter Binding Constants as a Function of Solution Conditions," Biochemistry, ( ) **19**, pp. 3504-3515.

Sweetser, *et al.*, Proc. Natl. Acad. Sci. USA, (1987) **84**, p. 1192.

Tanaka, K., Y. et al., "Heterogeneity of principal σ factor in Escherichia coli: the rpoS gene product, σ^{38}, is a second principal σ factor of RNA polymerase in

stationary-phase Escherichia coli," Proc. Natl. Acad. Sci. USA, (1993) **90**, pp. 3511-3515.

Wang, Y., et al., "Determinants for Escherichia coli RNA polymerase assembly within the β subunit," J. Mol. Biol., (1997) **270**, pp. 648-662.

Yura, T., et al., "Genetics of bacterial RNA polymerase," Annu. Rev. Genet., (1979) **13**, pp. 59-97.

Zillig, W., et al., "Function and reassembly of subunits of DNA-dependent RNA polymerase," In RNA Polymerase, (Cold Spring Harbor Laboratory Press, Cold Spring Harbor, NY.) (1976) pp. 101-125.

I claim:

1. A method of interfering with bacterial life cycle comprising the step of bringing bacterial cells into contact with a compound that blocks the binding of at least one protein subunit of RNAP to a second protein subunit of RNAP.

2. The method of Claim 1 wherein the compound blocks the binding of the σ-subunit to the β′-subunit.

3. The method of Claim 2 wherein the compound binds to β'-subunit amino acid residues 60 through 135.

4. The method of Claim 2 wherein the compound is or binds to an amino acid sequence selected from the group consisting of
RIFGPVKDYECLCGKYKRLKHRGVICEKCGVEVTQTKVRRERMGHIELASPT
AHIWFLKSLPSRIGLLLDMPLRDI (SEQ ID NO:4);
ARIFGPVKDHECLCGKYKRLKHRG (SEQ ID NO:5);
ICEKCGVEVTQTKVRRERMGHI (SEQ ID NO:6);
CHIWFLKSLPSRIGLLLDMPLRDIE (SEQ ID NO:7);
ECLCGKYKRLKHRGVICEKCGV (SEQ ID NO:8); and
CKVRRERMGHIELASPTAHIWFLKSL (SEQ ID NO:9).

5. The method of Claim 2 wherein the compound is a derivative of an amino acid sequence selected from the group consisting of
RIFGPVKDYECLCGKYKRLKHRGVICEKCGVEVTQTKVRRERMGHIELASPT
AHIWFLKSLPSRIGLLLDMPLRDI (SEQ ID NO:4);
ARIFGPVKDHECLCGKYKRLKHRG (SEQ ID NO:5);
ICEKCGVEVTQTKVRRERMGHI (SEQ ID NO:6);
CHIWFLKSLPSRIGLLLDMPLRDIE (SEQ ID NO:7);
ECLCGKYKRLKHRGVICEKCGV (SEQ ID NO:8); and
CKVRRERMGHIELASPTAHIWFLKSL (SEQ ID NO:9).

6. The method of Claim 1 wherein the compound blocks the binding of the α-subunit to the β′-subunit.

7. The method of Claim 6 wherein the compound binds to α-subunit amino acid residues 175 through 185 or 195 through 210.

8. The method of Claim 6 wherein the compound is or binds to an amino acid sequence selected from the group consisting of peptides Leu Val Asp Ala Cys Tyr Ser Pro Val Glu Arg Ile Ala Tyr Asn Val Glu Ala (SEQ. ID No. 21), Ala Ala Arg Val Glu Gln Arg Thr Asp Leu Asp Lys Leu Val Ile Glu Met Glu Thr Asn Gly Thr (SEQ. ID No. 22), and Val Glu Arg Ile Ala Tyr Asn Val Glu Ala Ala Arg Val Glu Gln Arg Thr Asp Leu Asp Lys Leu Val (SEQ. ID No. 23).

9. The method of Claim 6 wherein the compound is a derivative of an amino acid sequence selected from the group consisting of peptides Leu Val Asp Ala Cys Tyr Ser Pro Val Glu Arg Ile Ala Tyr Asn Val Glu Ala (SEQ. ID No. 21), Ala Ala Arg Val Glu Gln Arg Thr Asp Leu Asp Lys Leu Val Ile Glu Met Glu Thr Asn Gly Thr (SEQ. ID No. 22), and Val Glu Arg Ile Ala Tyr Asn Val Glu Ala Ala Arg Val Glu Gln Arg Thr Asp Leu Asp Lys Leu Val (SEQ. ID No. 23).

10. The method of Claim 1 wherein the compound blocks the binding of the α-subunit to the β-subunit.

11. The method of Claim 10 wherein the compound binds to α-subunit amino acid residues 30 through 55 or 61 through 76.

12. The method of Claim 10 wherein the compound is or binds to an amino acid sequence selected from the group consisting of peptides Pro Leu Glu Arg Gly Phe Gly His Thr Leu Gly Asn Ala Leu Arg Arg Ile Leu Leu Ser Met Pro Gly Cys (SEQ. ID No. 18), Cys Ala Val Thr Glu Val Glu Ile Asp Gly Val Leu His Glu Tyr Ser Thr Lys Glu Gly Val Gln Glu Asp Ile (SEQ. ID No. 19), and Asn Ala Leu Arg Arg Ile Leu Leu Ser Met Pro Gly Cys AlaVal Thr Glu Val Glu Ile Asp Gly Val Leu (SEQ. ID No. 20).

13. The method of Claim 10 wherein the compound is a derivative of an amino acid sequence selected from the group consisting of peptides Pro Leu Glu

Arg Gly Phe Gly His Thr Leu Gly Asn Ala Leu Arg Arg Ile Leu Leu Ser Met Pro Gly Cys (SEQ. ID No. 18), Cys Ala Val Thr Glu Val Glu Ile Asp Gly Val Leu His Glu Tyr Ser Thr Lys Glu Gly Val Gln Glu Asp Ile (SEQ. ID No. 19), and Asn Ala Leu Arg Arg Ile Leu Leu Ser Met Pro Gly Cys AlaVal Thr Glu Val Glu Ile Asp Gly Val Leu (SEQ. ID No. 20).

14. The method of Claim 10 wherein the compound binds to β-subunit amino acid residues 907 through 1246.

15. The method of Claim 10 wherein the compound is or binds to an amino acid sequence selected from the group consisting of peptides Thr His Leu Gly Met Ala Ala Lys Gly Ile Gly Asp Lys Ile Asn Ala Met Leu Lys Gln Gln Gln Glu Val (SEQ. ID No. 26), Ala Lys Leu Arg Glu Phe Ile Gln Arg Ala Tyr Asp Leu Gly Ala Asp Val Arg Gln Lys Val Asp Leu Ser (SEQ. ID No. 27), Gly Asp Lys Ile Asn Ala Met Leu Lys Gln Gln Gln Glu Val Ala Lys Leu Arg Glu Phe Ile Gln Arg Ala (SEQ. ID No. 28), Thr Phe Ser Asp Glu Glu Val Met Arg Leu Ala Glu Asn Leu Arg Lys Gly Met Pro Ieu Ala Thr Pro Val (SEQ. ID No. 29), Phe Asp Gly Ala Lys Glu Ala Glu Ile Lys Glu Leu Leu Lys Leu Gly Asp Leu Pro Thr Ser Gly Gln Ile (SEQ. ID No. 30), Arg Leu Tyr Asp Gly Arg Thr Gly Glu Gln Phe Glu Arg Pro Val Thr Val Gly Tyr Met Tyr Met Leu Lys (SEQ. ID No. 31), Arg Pro Val Thr Val Gly Tyr Met Tyr Met Leu Lys Leu Asn His Leu Val Asp Asp Lys Met His Ala Arg (SEQ. ID No. 32), Tyr Asp Leu Gly Ala Asp Val Arg Gln Lys Val Asp Leu Ser Thr Phe Ser Asp Glu Glu Val Met Arg Leu (SEQ. ID No. 33), Ala Glu Asn Leu Arg Lys Gly Met Pro Ieu Ala Thr Pro Val Phe Asp Gly Ala Lys Glu Ala Glu Ile Lys Glu Leu (SEQ. ID No. 34), Leu Lys Leu Gly Asp Leu Pro Thr Ser Gly Gln Ile Arg Leu Tyr Asp Gly Arg Thr (SEQ. ID No. 35), and Thr Gly Glu Gln Phe Glu Arg Pro Val Thr Val Gly Tyr Met Tyr Met Leu Lys Leu Asn His Leu (SEQ. ID No. 36).

16. The method of Claim 10 wherein the compound is a derivative of an amino acid sequence selected from the group consisting of peptides Thr His Leu Gly Met Ala Ala Lys Gly Ile Gly Asp Lys Ile Asn Ala Met Leu Lys Gln Gln Gln Glu Val (SEQ. ID No. 26). Ala Lys Leu Arg Glu Phe Ile Gln Arg Ala Tyr Asp Leu Gly Ala Asp Val Arg Gln Lys Val Asp Leu Ser (SEQ. ID No. 27). Gly Asp Lys Ile Asn

Ala Met Leu Lys Gln Gln Gln Glu Val Ala Lys Leu Arg Glu Phe Ile Gln Arg Ala (SEQ. ID No. 28), Thr Phe Ser Asp Glu Glu Val Met Arg Leu Ala Glu Asn Leu Arg Lys Gly Met Pro Ieu Ala Thr Pro Val (SEQ. ID No. 29), Phe Asp Gly Ala Lys Glu Ala Glu Ile Lys Glu Leu Leu Lys Leu Gly Asp Leu Pro Thr Ser Gly Gln Ile (SEQ. ID No. 30), Arg Leu Tyr Asp Gly Arg Thr Gly Glu Gln Phe Glu Arg Pro Val Thr Val Gly Tyr Met Tyr Met Leu Lys (SEQ. ID No. 31), Arg Pro Val Thr Val Gly Tyr Met Tyr Met Leu Lys Leu Asn His Leu Val Asp Asp Lys Met His Ala Arg (SEQ. ID No. 32), Tyr Asp Leu Gly Ala Asp Val Arg Gln Lys Val Asp Leu Ser Thr Phe Ser Asp Glu Glu Val Met Arg Leu (SEQ. ID No. 33), Ala Glu Asn Leu Arg Lys Gly Met Pro Ieu Ala Thr Pro Val Phe Asp Gly Ala Lys Glu Ala Glu Ile Lys Glu Leu (SEQ. ID No. 34), Leu Lys Leu Gly Asp Leu Pro Thr Ser Gly Gln Ile Arg Leu Tyr Asp Gly Arg Thr (SEQ. ID No. 35), and Thr Gly Glu Gln Phe Glu Arg Pro Val Thr Val Gly Tyr Met Tyr Met Leu Lys Leu Asn His Leu (SEQ. ID No. 36).

17. The method of Claim 1 wherein the compound blocks the binding of the β′-subunit to the β-subunit.

18. The method of Claim 17 wherein the compound binds to β-subunit amino acid residues 1247 through 1342.

19. The method of Claim 17 wherein the compound is or binds to an amino acid sequence selected from the group consisting of peptides Ser Thr Gly Ser Tyr Ser Leu Val Thr Gln Gln Pro Leu Gly Gly Lys Ala Gln Phe Gly (SEQ. ID No. 37), Gly Gln Arg Phe Gly Glu Met Glu Val Trp Ala Leu Glu Ala Tyr Gly Ala Ala Tyr Thr Leu Gln Glu Met (SEQ. ID No. 38), Met Leu Thr Val Lys Ser Asp Asp Val Asn Gly Arg Thr Lys Met Tyr Lys Asn Ile Val Asp Gly (SEQ. ID No. 39), Asn His Gln Met Glu Pro Gly Met Pro Glu Ser Phe Asn Val Leu Leu Lys Glu Ile Arg Ser Leu Gly (SEQ. ID No. 40), Met Pro Glu Ser Phe Asn Val Leu Leu Lys Glu Ile Arg Ser Leu Gly Ile Asn Ile Glu Leu Glu Asp Glu (SEQ. ID No. 41), Thr Gln Gln Pro Leu Gly Gly Lys Ala Gln Phe Gly Gly Gln Arg Phe Gly Glu Met Glu Val (SEQ. ID No. 42), Val Trp Ala Leu Glu Ala Tyr Gly Ala Ala Tyr Thr Leu Gln Glu Met Leu Thr Val Lys Ser Asp Asp Val (SEQ. ID No. 43), and Val Asn Gly Arg Thr Lys Met Tyr Lys Asn Ile Val Asp Giy Asn His Gln Met Glu Pro Gly (SEQ. ID No. 44).

20. The method of Claim 17 wherein the compound is a derivative of an amino acid sequence selected from the group consisting of peptides Ser Thr Gly Ser Tyr Ser Leu Val Thr Gln Gln Pro Leu Gly Gly Lys Ala Gln Phe Gly (SEQ. ID No. 37), Gly Gln Arg Phe Gly Glu Met Glu Val Trp Ala Leu Glu Ala Tyr Gly Ala Ala Tyr Thr Leu Gln Glu Met (SEQ. ID No. 38), Met Leu Thr Val Lys Ser Asp Asp Val Asn Gly Arg Thr Lys Met Tyr Lys Asn Ile Val Asp Gly (SEQ. ID No. 39), Asn His Gln Met Glu Pro Gly Met Pro Glu Ser Phe Asn Val Leu Leu Lys Glu Ile Arg Ser Leu Gly (SEQ. ID No. 40), Met Pro Glu Ser Phe Asn Val Leu Leu Lys Glu Ile Arg Ser Leu Gly Ile Asn Ile Glu Leu Glu Asp Glu (SEQ. ID No. 41), Thr Gln Gln Pro Leu Gly Gly Lys Ala Gln Phe Gly Gly Gln Arg Phe Gly Glu Met Glu Val (SEQ. ID No. 42), Val Trp Ala Leu Glu Ala Tyr Gly Ala Ala Tyr Thr Leu Gln Glu Met Leu Thr Val Lys Ser Asp Asp Val (SEQ. ID No. 43), and Val Asn Gly Arg Thr Lys Met Tyr Lys Asn Ile Val Asp Gly Asn His Gln Met Glu Pro Gly (SEQ. ID No. 44).

21. A method of interfering with bacterial life cycle comprising the step of bringing bacterial cells into contact with a compound that blocks the binding of the σ-subunit of RNAP to the RNAP core.

22. The method of Claim 21 wherein the compound binds to σ70-subunit amino acid residues 361 through 390.

23. The method of Claim 21 wherein the compound is or binds to an amino acid sequence selected from the group consisting of Ile Asn Arg Arg Met Ser Ile Gly Glu Ala Lys Ala Arg Arg Ala (SEQ. ID No. 48), Ala Lys Lys Glu Met Val Glu Ala Asn Leu Arg Leu Val Ile Ser Ile (SEQ. ID No. 49), and Met Ser Ile Gly Glu Ala Lys Ala Arg Arg Ala Lys Lys Glu Met Val Glu Ala (SEQ. ID No. 50).

24. The method of Claim 21 wherein the compound is a derivative of an amino acid sequence selected from the group consisting of Ile Asn Arg Arg Met Ser Ile Gly Glu Ala Lys Ala Arg Arg Ala (SEQ. ID No. 48), Ala Lys Lys Glu Met Val Glu Ala Asn Leu Arg Leu Val Ile Ser Ile (SEQ. ID No. 49), and Met Ser Ile Gly Glu Ala Lys Ala Arg Arg Ala Lys Lys Glu Met Val Glu Ala (SEQ. ID No. 50).

25. A method of interfering with bacterial life cycle comprising the step of bringing bacterial cells into contact with a compound that blocks nucleic acid binding to the β′-subunit of RNAP.

26. The method of Claim 25 wherein the compound binds to β′-subunit amino acid residues 1 through 61 or 298 through 330.

27. The method of Claim 25 wherein the compound is or binds to an amino acid sequence selected from the group consisting of Met Lys Asp Leu Leu Lys Phe Leu Lys Ala Gln Thr Lys Thr Glu Glu Phe Asp Ala Ile Lys Ile Ala (SEQ. ID No. 54), Ile Ala Leu Ala Ser Pro Asp Met Ile Arg Ser Trp Ser Phe Gly Glu Val Lys Lys Pro Glu Thr Ile (SEQ. ID No. 55), Thr Ile Asn Tyr Arg Thr Phe Lys Pro Glu Arg Asp Gly Leu Phe Cys Ala Arg Ile (SEQ. ID No. 56), Val Lys Lys Pro Glu Thr Ile Asn Tyr Arg Thr Phe Lys Pro Glu Arg Asp Gly Leu Phe Cys (SEQ. ID No. 57), Thr Glu Glu Phe Asp Ala Ile Lys Ile Ala Leu Ala Ser Pro Asp Met Ile Arg Ser Trp Ser Phe Gly Glu Val (SEQ. ID No. 58), Met Leu Gln Glu Ala Val Asp Ala Leu Leu Asp Asn Gly Arg Arg Gly Arg Ala (SEQ. ID No. 59), Ala Ile Thr Gly Ser Asn Lys Lys Pro Leu Lys Ser Leu Ala Asp Met (SEQ. ID No. 60), and Leu Asp Asn Gly Arg Arg Gly Arg Ala Ile Thr Gly Ser Asn Lys Lys Pro Leu Lys Ser Leu (SEQ. ID No. 61).

28. The method of Claim 21 wherein the compound is a derivative of an amino acid sequence selected from the group consisting of Met Lys Asp Leu Leu Lys Phe Leu Lys Ala Gln Thr Lys Thr Glu Glu Phe Asp Ala Ile Lys Ile Ala (SEQ. ID No. 54), Ile Ala Leu Ala Ser Pro Asp Met Ile Arg Ser Trp Ser Phe Gly Glu Val Lys Lys Pro Glu Thr Ile (SEQ. ID No. 55), Thr Ile Asn Tyr Arg Thr Phe Lys Pro Glu Arg Asp Gly Leu Phe Cys Ala Arg Ile (SEQ. ID No. 56), Val Lys Lys Pro Glu Thr Ile Asn Tyr Arg Thr Phe Lys Pro Glu Arg Asp Gly Leu Phe Cys (SEQ. ID No. 57), Thr Glu Glu Phe Asp Ala Ile Lys Ile Ala Leu Ala Ser Pro Asp Met Ile Arg Ser Trp Ser Phe Gly Glu Val (SEQ. ID No. 58), Met Leu Gln Glu Ala Val Asp Ala Leu Leu Asp Asn Gly Arg Arg Gly Arg Ala (SEQ. ID No. 59), Ala Ile Thr Gly Ser Asn Lys Lys Pro Leu Lys Ser Leu Ala Asp Met (SEQ. ID No. 60), and Leu Asp Asn Gly Arg Arg Gly Arg Ala Ile Thr Gly Ser Asn Lys Lys Pro Leu Lys Ser Leu (SEQ. ID No. 61).

29. A method of interfering with bacterial life cycle comprising the step of bringing bacterial cells into contact with a compound that blocks nucleic acid binding to the β-subunit of RNAP.

30. The method of Claim 29 wherein the compound binds to β-subunit amino acid residues 130 through 239 or 1230 through 1304.

31. The method of Claim 29 wherein the compound is or binds to an amino acid sequence selected from the group consisting of Met Thr Asp Asn Gly Thr Phe Val Ile Asn Gly Thr Glu Arg Val Ile Val Ser Gln Leu His Arg (SEQ. ID No. 66), Ser Pro Gly Val Phe Phe Asp Ser Asp Lys Gly Lys Thr His Ser Ser Gly Lys Val Leu Tyr Asn (SEQ. ID No. 67), Ala Arg Ile Ile Pro Tyr Arg Gly Ser Trp Leu Asp Phe Glu Phe Asp Pro Lys Asp Asn Leu Phe (SEQ. ID No. 68), Val Arg Ile Asp Arg Arg Arg Lys Leu Pro Ala Thr Ile Ile Leu Arg Ala Leu Asn Tyr Thr Thr (SEQ. ID No. 69), Glu Gln Ile Leu Asp Leu Phe Phe Glu Lys Val Ile Phe Glu Ile Arg Asp Asn Lys Leu Gln Met (SEQ. ID No. 70), Ser Trp Leu Asp Phe Glu Phe Asp Pro Lys Asp Asn Leu Phe Val Arg Ile Asp Arg Arg Arg Lys Leu Pro (SEQ. ID No. 71), Val Ser Gln Leu His Arg Ser Pro Gly Val Phe Phe Asp Ser Asp Lys Gly Lys Thr His Ser (SEQ. ID No. 72), Ser Gly Lys Val Leu Tyr Asn Ala Arg Ile Ile Pro Tyr Arg Gly Ser Trp Leu (SEQ. ID No. 73), Leu Asp Phe Glu Phe Asp Pro Lys Asp Asn Leu Phe Val Arg Ile Asp Arg Arg Arg Lys Leu Pro (SEQ. ID No. 74), Ala Thr Ile Ile Leu Arg Ala Leu Asn Tyr Thr Thr Glu Gln Ile Leu Asp Leu Phe Phe Glu Lys Val (SEQ. ID No. 75), Met Tyr Met Leu Lys Leu Asn His Leu Val Asp Asp Lys Met His Ala Arg Ser Thr Gly Ser Tyr Ser Leu Val (SEQ. ID No. 76), Thr Gln Gln Pro Leu Gly Gly Lys Ala Gln Phe Gly Gly Gln Arg Phe Gly Glu Met Glu Val Trp Ala Leu Glu (SEQ. ID No. 77), Ala Tyr Gly Ala Ala Tyr Thr Leu Gln Glu Met Leu Thr Val Lys Ser Asp Asp Val Asn Gly Arg Thr Lys Met (SEQ. ID No. 78), Met His Ala Arg Ser Thr Gly Ser Tyr Ser Leu Val Thr Gln Gln Pro Leu Gly Gly Lys Ala Gln Phe Gly (SEQ. ID No. 79)and Gly Gln Arg Phe Gly Glu Met Glu Val Trp Ala Leu Glu Ala Tyr Gly Ala Ala Tyr Thr Leu Gln Glu Met Leu (SEQ. ID No. 80).

32. The method of Claim 29 wherein the compound is a derivative of an amino acid sequence selected from the group consisting of Met Thr Asp Asn Gly Thr Phe Val Ile Asn Gly Thr Glu Arg Val Ile Val Ser Gln Leu His Arg (SEQ. ID No. 66), Ser Pro Gly Val Phe Phe Asp Ser Asp Lys Gly Lys Thr His Ser Ser Gly Lys Val Leu Tyr Asn (SEQ. ID No. 67), Ala Arg Ile Ile Pro Tyr Arg Gly Ser Trp Leu Asp Phe Glu Phe Asp Pro Lys Asp Asn Leu Phe (SEQ. ID No. 68), Val Arg Ile Asp Arg Arg Arg Lys Leu Pro Ala Thr Ile Ile Leu Arg Ala Leu Asn Tyr Thr Thr (SEQ. ID No. 69), Glu Gln Ile Leu Asp Leu Phe Phe Glu Lys Val Ile Phe Glu Ile Arg Asp Asn Lys Leu Gln Met (SEQ. ID No. 70), Ser Trp Leu Asp Phe Glu Phe Asp Pro Lys Asp Asn Leu Phe Val Arg Ile Asp Arg Arg Arg Lys Leu Pro (SEQ. ID No. 71), Val Ser Gln Leu His Arg Ser Pro Gly Val Phe Phe Asp Ser Asp Lys Gly Lys Thr His Ser (SEQ. ID No. 72), Ser Gly Lys Val Leu Tyr Asn Ala Arg Ile Ile Pro Tyr Arg Gly Ser Trp Leu (SEQ. ID No. 73), Leu Asp Phe Glu Phe Asp Pro Lys Asp Asn Leu Phe Val Arg Ile Asp Arg Arg Arg Lys Leu Pro (SEQ. ID No. 74), Ala Thr Ile Ile Leu Arg Ala Leu Asn Tyr Thr Thr Glu Gln Ile Leu Asp Leu Phe Phe Glu Lys Val (SEQ. ID No. 75), Met Tyr Met Leu Lys Leu Asn His Leu Val Asp Asp Lys Met His Ala Arg Ser Thr Gly Ser Tyr Ser Leu Val (SEQ. ID No. 76), Thr Gln Gln Pro Leu Gly Gly Lys Ala Gln Phe Gly Gly Gln Arg Phe Gly Glu Met Glu Val Trp Ala Leu Glu (SEQ. ID No. 77), Ala Tyr Gly Ala Ala Tyr Thr Leu Gln Glu Met Leu Thr Val Lys Ser Asp Asp Val Asn Gly Arg Thr Lys Met (SEQ. ID No. 78), Met His Ala Arg Ser Thr Gly Ser Tyr Ser Leu Val Thr Gln Gln Pro Leu Gly Gly Lys Ala Gln Phe Gly (SEQ. ID No. 79)and Gly Gln Arg Phe Gly Glu Met Glu Val Trp Ala Leu Glu Ala Tyr Gly Ala Ala Tyr Thr Leu Gln Glu Met Leu (SEQ. ID No. 80).

33. A method of inhibiting bacterial growth comprising the step of bringing bacterial cells into contact with a compound that blocks the binding of at least one protein subunit of RNAP to a second protein subunit of RNAP.

34. The method of Claim 33 wherein the compound blocks the binding of the σ-subunit to the β'-subunit.

35. The method of Claim 34 wherein the compound binds to β'-subunit amino acid residues 60 through 135.

36. The method of Claim 34 wherein the compound is or binds to an amino acid sequence selected from the group consisting of
RIFGPVKDYECLCGKYKRLKHRGVICEKCGVEVTQTKVRRERMGHIELASPT
AHIWFLKSLPSRIGLLLDMPLRDI (SEQ ID NO:4);
ARIFGPVKDHECLCGKYKRLKHRG (SEQ ID NO:5);
ICEKCGVEVTQTKVRRERMGHI (SEQ ID NO:6);
CHIWFLKSLPSRIGLLLDMPLRDIE (SEQ ID NO:7);
ECLCGKYKRLKHRGVICEKCGV (SEQ ID NO:8); and
CKVRRERMGHIELASPTAHIWFLKSL (SEQ ID NO:9).

37. The method of Claim 34 wherein the compound is a derivative of an amino acid sequence selected from the group consisting of
RIFGPVKDYECLCGKYKRLKHRGVICEKCGVEVTQTKVRRERMGHIELASPT
AHIWFLKSLPSRIGLLLDMPLRDI (SEQ ID NO:4);
ARIFGPVKDHECLCGKYKRLKHRG (SEQ ID NO:5);
ICEKCGVEVTQTKVRRERMGHI (SEQ ID NO:6);
CHIWFLKSLPSRIGLLLDMPLRDIE (SEQ ID NO:7);
ECLCGKYKRLKHRGVICEKCGV (SEQ ID NO:8); and
CKVRRERMGHIELASPTAHIWFLKSL (SEQ ID NO:9).

38. The method of Claim 33 wherein the compound blocks the binding of the α-subunit to the β′-subunit.

39. The method of Claim 38 wherein the compound binds to α-subunit amino acid residues 175 through 185 or 195 through 210.

40. The method of Claim 38 wherein the compound is or binds to an amino acid sequence selected from the group consisting of peptides Leu Val Asp Ala Cys Tyr Ser Pro Val Glu Arg Ile Ala Tyr Asn Val Glu Ala (SEQ. ID No. 21), Ala Ala Arg Val Glu Gln Arg Thr Asp Leu Asp Lys Leu Val Ile Glu Met Glu Thr Asn Gly

Thr (SEQ. ID No. 22), and Val Glu Arg Ile Ala Tyr Asn Val Glu Ala Ala Arg Val Glu Gln Arg Thr Asp Leu Asp Lys Leu Val (SEQ. ID No. 23).

41. The method of Claim 38 wherein the compound is a derivative of an amino acid sequence selected from the group consisting of peptides Leu Val Asp Ala Cys Tyr Ser Pro Val Glu Arg Ile Ala Tyr Asn Val Glu Ala (SEQ. ID No. 21), Ala Ala Arg Val Glu Gln Arg Thr Asp Leu Asp Lys Leu Val Ile Glu Met Glu Thr Asn Gly Thr (SEQ. ID No. 22), and Val Glu Arg Ile Ala Tyr Asn Val Glu Ala Ala Arg Val Glu Gln Arg Thr Asp Leu Asp Lys Leu Val (SEQ. ID No. 23).

42. The method of Claim 33 wherein the compound blocks the binding of the α-subunit to the β-subunit.

43. The method of Claim 42 wherein the compound binds to α-subunit amino acid residues 30 through 55 or 61 through 76.

44. The method of Claim 42 wherein the compound is or binds to an amino acid sequence selected from the group consisting of peptides Pro Leu Glu Arg Gly Phe Gly His Thr Leu Gly Asn Ala Leu Arg Arg Ile Leu Leu Ser Met Pro Gly Cys (SEQ. ID No. 18), Cys Ala Val Thr Glu Val Glu Ile Asp Gly Val Leu His Glu Tyr Ser Thr Lys Glu Gly Val Gln Glu Asp Ile (SEQ. ID No. 19), and Asn Ala Leu Arg Arg Ile Leu Leu Ser Met Pro Gly Cys AlaVal Thr Glu Val Glu Ile Asp Gly Val Leu (SEQ. ID No. 20).

45. The method of Claim 42 wherein the compound is a derivative of an amino acid sequence selected from the group consisting of peptides Pro Leu Glu Arg Gly Phe Gly His Thr Leu Gly Asn Ala Leu Arg Arg Ile Leu Leu Ser Met Pro Gly Cys (SEQ. ID No. 18), Cys Ala Val Thr Glu Val Glu Ile Asp Gly Val Leu His Glu Tyr Ser Thr Lys Glu Gly Val Gln Glu Asp Ile (SEQ. ID No. 19), and Asn Ala Leu Arg Arg Ile Leu Leu Ser Met Pro Gly Cys AlaVal Thr Glu Val Glu Ile Asp Gly Val Leu (SEQ. ID No. 20).

46. The method of Claim 42 wherein the compound binds to β-subunit amino acid residues 907 through 1246.

47. The method of Claim 42 wherein the compound is or binds to an amino acid sequence selected from the group consisting of peptides Thr His Leu Gly Met Ala Ala Lys Gly Ile Gly Asp Lys Ile Asn Ala Met Leu Lys Gln Gln Gln Glu Val (SEQ. ID No. 26), Ala Lys Leu Arg Glu Phe Ile Gln Arg Ala Tyr Asp Leu Gly Ala Asp Val Arg Gln Lys Val Asp Leu Ser (SEQ. ID No. 27), Gly Asp Lys Ile Asn Ala Met Leu Lys Gln Gln Gln Glu Val Ala Lys Leu Arg Glu Phe Ile Gln Arg Ala (SEQ. ID No. 28), Thr Phe Ser Asp Glu Glu Val Met Arg Leu Ala Glu Asn Leu Arg Lys Gly Met Pro Ieu Ala Thr Pro Val (SEQ. ID No. 29), Phe Asp Gly Ala Lys Glu Ala Glu Ile Lys Glu Leu Leu Lys Leu Gly Asp Leu Pro Thr Ser Gly Gln Ile (SEQ. ID No. 30), Arg Leu Tyr Asp Gly Arg Thr Gly Glu Gln Phe Glu Arg Pro Val Thr Val Gly Tyr Met Tyr Met Leu Lys (SEQ. ID No. 31), Arg Pro Val Thr Val Gly Tyr Met Tyr Met Leu Lys Leu Asn His Leu Val Asp Asp Lys Met His Ala Arg (SEQ. ID No. 32), Tyr Asp Leu Gly Ala Asp Val Arg Gln Lys Val Asp Leu Ser Thr Phe Ser Asp Glu Glu Val Met Arg Leu (SEQ. ID No. 33), Ala Glu Asn Leu Arg Lys Gly Met Pro Ieu Ala Thr Pro Val Phe Asp Gly Ala Lys Glu Ala Glu Ile Lys Glu Leu (SEQ. ID No. 34), Leu Lys Leu Gly Asp Leu Pro Thr Ser Gly Gln Ile Arg Leu Tyr Asp Gly Arg Thr (SEQ. ID No. 35), and Thr Gly Glu Gln Phe Glu Arg Pro Val Thr Val Gly Tyr Met Tyr Met Leu Lys Leu Asn His Leu (SEQ. ID No. 36).

48. The method of Claim 42 wherein the compound is a derivative of an amino acid sequence selected from the group consisting of peptides Thr His Leu Gly Met Ala Ala Lys Gly Ile Gly Asp Lys Ile Asn Ala Met Leu Lys Gln Gln Gln Glu Val (SEQ. ID No. 26), Ala Lys Leu Arg Glu Phe Ile Gln Arg Ala Tyr Asp Leu Gly Ala Asp Val Arg Gln Lys Val Asp Leu Ser (SEQ. ID No. 27), Gly Asp Lys Ile Asn Ala Met Leu Lys Gln Gln Gln Glu Val Ala Lys Leu Arg Glu Phe Ile Gln Arg Ala (SEQ. ID No. 28), Thr Phe Ser Asp Glu Glu Val Met Arg Leu Ala Glu Asn Leu Arg Lys Gly Met Pro Ieu Ala Thr Pro Val (SEQ. ID No. 29), Phe Asp Gly Ala Lys Glu Ala Glu Ile Lys Glu Leu Leu Lys Leu Gly Asp Leu Pro Thr Ser Gly Gln Ile (SEQ. ID No. 30), Arg Leu Tyr Asp Gly Arg Thr Gly Glu Gln Phe Glu Arg Pro Val Thr Val Gly Tyr Met Tyr Met Leu Lys (SEQ. ID No. 31), Arg Pro Val Thr Val Gly Tyr Met Tyr Met Leu Lys Leu Asn His Leu Val Asp Asp Lys Met His Ala Arg (SEQ. ID No.

32), Tyr Asp Leu Gly Ala Asp Val Arg Gln Lys Val Asp Leu Ser Thr Phe Ser Asp Glu Glu Val Met Arg Leu (SEQ. ID No. 33), Ala Glu Asn Leu Arg Lys Gly Met Pro Ieu Ala Thr Pro Val Phe Asp Gly Ala Lys Glu Ala Glu Ile Lys Glu Leu (SEQ. ID No. 34). Leu Lys Leu Gly Asp Leu Pro Thr Ser Gly Gln Ile Arg Leu Tyr Asp Gly Arg Thr (SEQ. ID No. 35), and Thr Gly Glu Gln Phe Glu Arg Pro Val Thr Val Gly Tyr Met Tyr Met Leu Lys Leu Asn His Leu (SEQ. ID No. 36).

49. The method of Claim 33 wherein the compound blocks the binding of the β′-subunit to the β-subunit.

50. The method of Claim 49 wherein the compound binds to β-subunit amino acid residues 1247 through 1342.

51. The method of Claim 49 wherein the compound is or binds to an amino acid sequence selected from the group consisting of peptides Ser Thr Gly Ser Tyr Ser Leu Val Thr Gln Gln Pro Leu Gly Gly Lys Ala Gln Phe Gly (SEQ. ID No. 37), Gly Gln Arg Phe Gly Glu Met Glu Val Trp Ala Leu Glu Ala Tyr Gly Ala Ala Tyr Thr Leu Gln Glu Met (SEQ. ID No. 38), Met Leu Thr Val Lys Ser Asp Asp Val Asn Gly Arg Thr Lys Met Tyr Lys Asn Ile Val Asp Gly (SEQ. ID No. 39), Asn His Gln Met Glu Pro Gly Met Pro Glu Ser Phe Asn Val Leu Leu Lys Glu Ile Arg Ser Leu Gly (SEQ. ID No. 40), Met Pro Glu Ser Phe Asn Val Leu Leu Lys Glu Ile Arg Ser Leu Gly Ile Asn Ile Glu Leu Glu Asp Glu (SEQ. ID No. 41), Thr Gln Gln Pro Leu Gly Gly Lys Ala Gln Phe Gly Gly Gln Arg Phe Gly Glu Met Glu Val (SEQ. ID No. 42), Val Trp Ala Leu Glu Ala Tyr Gly Ala Ala Tyr Thr Leu Gln Glu Met Leu Thr Val Lys Ser Asp Asp Val (SEQ. ID No. 43), and Val Asn Gly Arg Thr Lys Met Tyr Lys Asn Ile Val Asp Gly Asn His Gln Met Glu Pro Gly (SEQ. ID No. 44).

52. The method of Claim 49 wherein the compound is a derivative of an amino acid sequence selected from the group consisting of peptides Ser Thr Gly Ser Tyr Ser Leu Val Thr Gln Gln Pro Leu Gly Gly Lys Ala Gln Phe Gly (SEQ. ID No. 37), Gly Gln Arg Phe Gly Glu Met Glu Val Trp Ala Leu Glu Ala Tyr Gly Ala Ala Tyr Thr Leu Gln Glu Met (SEQ. ID No. 38), Met Leu Thr Val Lys Ser Asp Asp Val Asn Gly Arg Thr Lys Met Tyr Lys Asn Ile Val Asp Gly (SEQ. ID No. 39), Asn His

Gln Met Glu Pro Gly Met Pro Glu Ser Phe Asn Val Leu Leu Lys Glu Ile Arg Ser Leu Gly (SEQ. ID No. 40), Met Pro Glu Ser Phe Asn Val Leu Leu Lys Glu Ile Arg Ser Leu Gly Ile Asn Ile Glu Leu Glu Asp Glu (SEQ. ID No. 41), Thr Gln Gln Pro Leu Gly Gly Lys Ala Gln Phe Gly Gly Gln Arg Phe Gly Glu Met Glu Val (SEQ. ID No. 42), Val Trp Ala Leu Glu Ala Tyr Gly Ala Ala Tyr Thr Leu Gln Glu Met Leu Thr Val Lys Ser Asp Asp Val (SEQ. ID No. 43), and Val Asn Gly Arg Thr Lys Met Tyr Lys Asn Ile Val Asp Gly Asn His Gln Met Glu Pro Gly (SEQ. ID No. 44).

53. A method of inhibiting bacterial growth comprising the step of bringing bacterial cells into contact with a compound that blocks the binding of the σ-subunit of RNAP to the RNAP core.

54. The method of Claim 53 wherein the compound binds to σ70-subunit amino acid residues 361 through 390.

55. The method of Claim 53 wherein the compound is or binds to an amino acid sequence selected from the group consisting of Ile Asn Arg Arg Met Ser Ile Gly Glu Ala Lys Ala Arg Arg Ala (SEQ. ID No. 48), Ala Lys Lys Glu Met Val Glu Ala Asn Leu Arg Leu Val Ile Ser Ile (SEQ. ID No. 49), and Met Ser Ile Gly Glu Ala Lys Ala Arg Arg Ala Lys Lys Glu Met Val Glu Ala (SEQ. ID No. 50).

56. The method of Claim 53 wherein the compound is a derivative of an amino acid sequence selected from the group consisting of Ile Asn Arg Arg Met Ser Ile Gly Glu Ala Lys Ala Arg Arg Ala (SEQ. ID No. 48), Ala Lys Lys Glu Met Val Glu Ala Asn Leu Arg Leu Val Ile Ser Ile (SEQ. ID No. 49), and Met Ser Ile Gly Glu Ala Lys Ala Arg Arg Ala Lys Lys Glu Met Val Glu Ala (SEQ. ID No. 50).

57. A method of inhibiting bacterial growth comprising the step of bringing bacterial cells into contact with a compound that blocks nucleic acid binding to the β'-subunit of RNAP.

58. The method of Claim 57 wherein the compound binds to β'-subunit amino acid residues 1 through 61 or 298 through 330.

59. The method of Claim 57 wherein the compound is or binds to an amino acid sequence selected from the group consisting of Met Lys Asp Leu Leu Lys Phe Leu Lys Ala Gln Thr Lys Thr Glu Glu Phe Asp Ala Ile Lys Ile Ala (SEQ. ID No. 54). Ile Ala Leu Ala Ser Pro Asp Met Ile Arg Ser Trp Ser Phe Gly Glu Val Lys Lys Pro Glu Thr Ile (SEQ. ID No. 55), Thr Ile Asn Tyr Arg Thr Phe Lys Pro Glu Arg Asp Gly Leu Phe Cys Ala Arg Ile (SEQ. ID No. 56), Val Lys Lys Pro Glu Thr Ile Asn Tyr Arg Thr Phe Lys Pro Glu Arg Asp Gly Leu Phe Cys (SEQ. ID No. 57), Thr Glu Glu Phe Asp Ala Ile Lys Ile Ala Leu Ala Ser Pro Asp Met Ile Arg Ser Trp Ser Phe Gly Glu Val (SEQ. ID No. 58), Met Leu Gln Glu Ala Val Asp Ala Leu Leu Asp Asn Gly Arg Arg Gly Arg Ala (SEQ. ID No. 59), Ala Ile Thr Gly Ser Asn Lys Lys Pro Leu Lys Ser Leu Ala Asp Met (SEQ. ID No. 60), and Leu Asp Asn Gly Arg Arg Gly Arg Ala Ile Thr Gly Ser Asn Lys Lys Pro Leu Lys Ser Leu (SEQ. ID No. 61).

60. The method of Claim 57 wherein the compound is a derivative of an amino acid sequence selected from the group consisting of Met Lys Asp Leu Leu Lys Phe Leu Lys Ala Gln Thr Lys Thr Glu Glu Phe Asp Ala Ile Lys Ile Ala (SEQ. ID No. 54), Ile Ala Leu Ala Ser Pro Asp Met Ile Arg Ser Trp Ser Phe Gly Glu Val Lys Lys Pro Glu Thr Ile (SEQ. ID No. 55), Thr Ile Asn Tyr Arg Thr Phe Lys Pro Glu Arg Asp Gly Leu Phe Cys Ala Arg Ile (SEQ. ID No. 56), Val Lys Lys Pro Glu Thr Ile Asn Tyr Arg Thr Phe Lys Pro Glu Arg Asp Gly Leu Phe Cys (SEQ. ID No. 57), Thr Glu Glu Phe Asp Ala Ile Lys Ile Ala Leu Ala Ser Pro Asp Met Ile Arg Ser Trp Ser Phe Gly Glu Val (SEQ. ID No. 58), Met Leu Gln Glu Ala Val Asp Ala Leu Leu Asp Asn Gly Arg Arg Gly Arg Ala (SEQ. ID No. 59), Ala Ile Thr Gly Ser Asn Lys Lys Pro Leu Lys Ser Leu Ala Asp Met (SEQ. ID No. 60), and Leu Asp Asn Gly Arg Arg Gly Arg Ala Ile Thr Gly Ser Asn Lys Lys Pro Leu Lys Ser Leu (SEQ. ID No. 61).

61. A method of inhibiting bacterial growth comprising the step of bringing bacterial cells into contact with a compound that blocks nucleic acid binding to the β-subunit of RNAP.

62. The method of Claim 61 wherein the compound binds to β-subunit amino acid residues 130 through 239 or 1230 through 1304.

63. The method of Claim 61 wherein the compound is or binds to an amino acid sequence selected from the group consisting of Met Thr Asp Asn Gly Thr Phe Val Ile Asn Gly Thr Glu Arg Val Ile Val Ser Gln Leu His Arg (SEQ. ID No. 66), Ser Pro Gly Val Phe Phe Asp Ser Asp Lys Gly Lys Thr His Ser Ser Gly Lys Val Leu Tyr Asn (SEQ. ID No. 67), Ala Arg Ile Ile Pro Tyr Arg Gly Ser Trp Leu Asp Phe Glu Phe Asp Pro Lys Asp Asn Leu Phe (SEQ. ID No. 68), Val Arg Ile Asp Arg Arg Arg Lys Leu Pro Ala Thr Ile Ile Leu Arg Ala Leu Asn Tyr Thr Thr (SEQ. ID No. 69), Glu Gln Ile Leu Asp Leu Phe Phe Glu Lys Val Ile Phe Glu Ile Arg Asp Asn Lys Leu Gln Met (SEQ. ID No. 70), Ser Trp Leu Asp Phe Glu Phe Asp Pro Lys Asp Asn Leu Phe Val Arg Ile Asp Arg Arg Arg Lys Leu Pro (SEQ. ID No. 71), Val Ser Gln Leu His Arg Ser Pro Gly Val Phe Phe Asp Ser Asp Lys Gly Lys Thr His Ser (SEQ. ID No. 72), Ser Gly Lys Val Leu Tyr Asn Ala Arg Ile Ile Pro Tyr Arg Gly Ser Trp Leu (SEQ. ID No. 73), Leu Asp Phe Glu Phe Asp Pro Lys Asp Asn Leu Phe Val Arg Ile Asp Arg Arg Arg Lys Leu Pro (SEQ. ID No. 74), Ala Thr Ile Ile Leu Arg Ala Leu Asn Tyr Thr Thr Glu Gln Ile Leu Asp Leu Phe Phe Glu Lys Val (SEQ. ID No. 75), Met Tyr Met Leu Lys Leu Asn His Leu Val Asp Asp Lys Met His Ala Arg Ser Thr Gly Ser Tyr Ser Leu Val (SEQ. ID No. 76), Thr Gln Gln Pro Leu Gly Gly Lys Ala Gln Phe Gly Gly Gln Arg Phe Gly Glu Met Glu Val Trp Ala Leu Glu (SEQ. ID No. 77), Ala Tyr Gly Ala Ala Tyr Thr Leu Gln Glu Met Leu Thr Val Lys Ser Asp Asp Val Asn Gly Arg Thr Lys Met (SEQ. ID No. 78), Met His Ala Arg Ser Thr Gly Ser Tyr Ser Leu Val Thr Gln Gln Pro Leu Gly Gly Lys Ala Gln Phe Gly (SEQ. ID No. 79)and Gly Gln Arg Phe Gly Glu Met Glu Val Trp Ala Leu Glu Ala Tyr Gly Ala Ala Tyr Thr Leu Gln Glu Met Leu (SEQ. ID No. 80).

64. The method of Claim 61 wherein the compound is a derivative of an amino acid sequence selected from the group consisting of Met Thr Asp Asn Gly Thr Phe Val Ile Asn Gly Thr Glu Arg Val Ile Val Ser Gln Leu His Arg (SEQ. ID No. 66), Ser Pro Gly Val Phe Phe Asp Ser Asp Lys Gly Lys Thr His Ser Ser Gly Lys Val Leu Tyr Asn (SEQ. ID No. 67), Ala Arg Ile Ile Pro Tyr Arg Gly Ser Trp Leu Asp Phe Glu Phe Asp Pro Lys Asp Asn Leu Phe (SEQ. ID No. 68), Val Arg Ile Asp Arg Arg Arg Lys Leu Pro Ala Thr Ile Ile Leu Arg Ala Leu Asn Tyr Thr Thr (SEQ. ID No. 69),

Glu Gln Ile Leu Asp Leu Phe Phe Glu Lys Val Ile Phe Glu Ile Arg Asp Asn Lys Leu Gln Met (SEQ. ID No. 70), Ser Trp Leu Asp Phe Glu Phe Asp Pro Lys Asp Asn Leu Phe Val Arg Ile Asp Arg Arg Arg Lys Leu Pro (SEQ. ID No. 71), Val Ser Gln Leu His Arg Ser Pro Gly Val Phe Phe Asp Ser Asp Lys Gly Lys Thr His Ser (SEQ. ID No. 72), Ser Gly Lys Val Leu Tyr Asn Ala Arg Ile Ile Pro Tyr Arg Gly Ser Trp Leu (SEQ. ID No. 73), Leu Asp Phe Glu Phe Asp Pro Lys Asp Asn Leu Phe Val Arg Ile Asp Arg Arg Arg Lys Leu Pro (SEQ. ID No. 74), Ala Thr Ile Ile Leu Arg Ala Leu Asn Tyr Thr Thr Glu Gln Ile Leu Asp Leu Phe Phe Glu Lys Val (SEQ. ID No. 75), Met Tyr Met Leu Lys Leu Asn His Leu Val Asp Asp Lys Met His Ala Arg Ser Thr Gly Ser Tyr Ser Leu Val (SEQ. ID No. 76), Thr Gln Gln Pro Leu Gly Gly Lys Ala Gln Phe Gly Gly Gln Arg Phe Gly Glu Met Glu Val Trp Ala Leu Glu (SEQ. ID No. 77), Ala Tyr Gly Ala Ala Tyr Thr Leu Gln Glu Met Leu Thr Val Lys Ser Asp Asp Val Asn Gly Arg Thr Lys Met (SEQ. ID No. 78), Met His Ala Arg Ser Thr Gly Ser Tyr Ser Leu Val Thr Gln Gln Pro Leu Gly Gly Lys Ala Gln Phe Gly (SEQ. ID No. 79)and Gly Gln Arg Phe Gly Glu Met Glu Val Trp Ala Leu Glu Ala Tyr Gly Ala Ala Tyr Thr Leu Gln Glu Met Leu (SEQ. ID No. 80).

65. A method of killing bacterial cells comprising the step of bringing bacterial cells into contact with a compound that blocks the binding of at least one protein subunit of RNAP to a second protein subunit of RNAP.

66. The method of Claim 65 wherein the compound blocks the binding of the σ-subunit to the β′-subunit.

67. The method of Claim 66 wherein the compound binds to β'-subunit amino acid residues 60 through 135.

68. The method of Claim 66 wherein the compound is or binds to an amino acid sequence selected from the group consisting of
RIFGPVKDYECLCGKYKRLKHRGVICEKCGVEVTQTKVRRERMGHIELASPT
AHIWFLKSLPSRIGLLLDMPLRDI (SEQ ID NO:4);
ARIFGPVKDHECLCGKYKRLKHRG (SEQ ID NO:5);
ICEKCGVEVTQTKVRRERMGHI (SEQ ID NO:6);

CHIWFLKSLPSRIGLLLDMPLRDIE (SEQ ID NO:7);
ECLCGKYKRLKHRGVICEKCGV (SEQ ID NO:8); and
CKVRRERMGHIELASPTAHIWFLKSL (SEQ ID NO:9).

69. The method of Claim 66 wherein the compound is a derivative of an amino acid sequence selected from the group consisting of
RIFGPVKDYECLCGKYKRLKHRGVICEKCGVEVTQTKVRRERMGHIELASPT
AHIWFLKSLPSRIGLLLDMPLRDI (SEQ ID NO:4);
ARIFGPVKDHECLCGKYKRLKHRG (SEQ ID NO:5);
ICEKCGVEVTQTKVRRERMGHI (SEQ ID NO:6);
CHIWFLKSLPSRIGLLLDMPLRDIE (SEQ ID NO:7);
ECLCGKYKRLKHRGVICEKCGV (SEQ ID NO:8); and
CKVRRERMGHIELASPTAHIWFLKSL (SEQ ID NO:9).

70. The method of Claim 65 wherein the compound blocks the binding of the α-subunit to the β′-subunit.

71. The method of Claim 70 wherein the compound binds to α-subunit amino acid residues 175 through 185 or 195 through 210.

72. The method of Claim 70 wherein the compound is or binds to an amino acid sequence selected from the group consisting of peptides Leu Val Asp Ala Cys Tyr Ser Pro Val Glu Arg Ile Ala Tyr Asn Val Glu Ala (SEQ. ID No. 21), Ala Ala Arg Val Glu Gln Arg Thr Asp Leu Asp Lys Leu Val Ile Glu Met Glu Thr Asn Gly Thr (SEQ. ID No. 22), and Val Glu Arg Ile Ala Tyr Asn Val Glu Ala Ala Arg Val Glu Gln Arg Thr Asp Leu Asp Lys Leu Val (SEQ. ID No. 23).

73. The method of Claim 70 wherein the compound is a derivative of an amino acid sequence selected from the group consisting of peptides Leu Val Asp Ala Cys Tyr Ser Pro Val Glu Arg Ile Ala Tyr Asn Val Glu Ala (SEQ. ID No. 21), Ala Ala Arg Val Glu Gln Arg Thr Asp Leu Asp Lys Leu Val Ile Glu Met Glu Thr Asn Gly Thr (SEQ. ID No. 22), and Val Glu Arg Ile Ala Tyr Asn Val Glu Ala Ala Arg Val Glu Gln Arg Thr Asp Leu Asp Lys Leu Val (SEQ. ID No. 23).

74. The method of Claim 65 wherein the compound blocks the binding of the α-subunit to the β-subunit.

75. The method of Claim 74 wherein the compound binds to α-subunit amino acid residues 30 through 55 or 61 through 76.

76. The method of Claim 74 wherein the compound is or binds to an amino acid sequence selected from the group consisting of peptides Pro Leu Glu Arg Gly Phe Gly His Thr Leu Gly Asn Ala Leu Arg Arg Ile Leu Leu Ser Met Pro Gly Cys (SEQ. ID No. 18), Cys Ala Val Thr Glu Val Glu Ile Asp Gly Val Leu His Glu Tyr Ser Thr Lys Glu Gly Val Gln Glu Asp Ile (SEQ. ID No. 19), and Asn Ala Leu Arg Arg Ile Leu Leu Ser Met Pro Gly Cys AlaVal Thr Glu Val Glu Ile Asp Gly Val Leu (SEQ. ID No. 20).

77. The method of Claim 74 wherein the compound is a derivative of an amino acid sequence selected from the group consisting of peptides Pro Leu Glu Arg Gly Phe Gly His Thr Leu Gly Asn Ala Leu Arg Arg Ile Leu Leu Ser Met Pro Gly Cys (SEQ. ID No. 18), Cys Ala Val Thr Glu Val Glu Ile Asp Gly Val Leu His Glu Tyr Ser Thr Lys Glu Gly Val Gln Glu Asp Ile (SEQ. ID No. 19), and Asn Ala Leu Arg Arg Ile Leu Leu Ser Met Pro Gly Cys AlaVal Thr Glu Val Glu Ile Asp Gly Val Leu (SEQ. ID No. 20).

78. The method of Claim 74 wherein the compound binds to β amino acid residues 907 through 1246.

79. The method of Claim 74 wherein the compound is or binds to an amino acid sequence selected from the group consisting of peptides Thr His Leu Gly Met Ala Ala Lys Gly Ile Gly Asp Lys Ile Asn Ala Met Leu Lys Gln Gln Gln Glu Val (SEQ. ID No. 26), Ala Lys Leu Arg Glu Phe Ile Gln Arg Ala Tyr Asp Leu Gly Ala Asp Val Arg Gln Lys Val Asp Leu Ser (SEQ. ID No. 27), Gly Asp Lys Ile Asn Ala Met Leu Lys Gln Gln Gln Glu Val Ala Lys Leu Arg Glu Phe Ile Gln Arg Ala (SEQ. ID No. 28), Thr Phe Ser Asp Glu Glu Val Met Arg Leu Ala Glu Asn Leu Arg Lys Gly Met Pro Ieu Ala Thr Pro Val (SEQ. ID No. 29), Phe Asp Gly Ala Lys Glu Ala Glu Ile

Lys Glu Leu Leu Lys Leu Gly Asp Leu Pro Thr Ser Gly Gln Ile (SEQ. ID No. 30), Arg Leu Tyr Asp Gly Arg Thr Gly Glu Gln Phe Glu Arg Pro Val Thr Val Gly Tyr Met Tyr Met Leu Lys (SEQ. ID No. 31). Arg Pro Val Thr Val Gly Tyr Met Tyr Met Leu Lys Leu Asn His Leu Val Asp Asp Lys Met His Ala Arg (SEQ. ID No. 32), Tyr Asp Leu Gly Ala Asp Val Arg Gln Lys Val Asp Leu Ser Thr Phe Ser Asp Glu Glu Val Met Arg Leu (SEQ. ID No. 33), Ala Glu Asn Leu Arg Lys Gly Met Pro Ieu Ala Thr Pro Val Phe Asp Gly Ala Lys Glu Ala Glu Ile Lys Glu Leu (SEQ. ID No. 34), Leu Lys Leu Gly Asp Leu Pro Thr Ser Gly Gln Ile Arg Leu Tyr Asp Gly Arg Thr (SEQ. ID No. 35), and Thr Gly Glu Gln Phe Glu Arg Pro Val Thr Val Gly Tyr Met Tyr Met Leu Lys Leu Asn His Leu (SEQ. ID No. 36).

80. The method of Claim 74 wherein the compound is a derivative of an amino acid sequence selected from the group consisting of peptides Thr His Leu Gly Met Ala Ala Lys Gly Ile Gly Asp Lys Ile Asn Ala Met Leu Lys Gln Gln Gln Glu Val (SEQ. ID No. 26), Ala Lys Leu Arg Glu Phe Ile Gln Arg Ala Tyr Asp Leu Gly Ala Asp Val Arg Gln Lys Val Asp Leu Ser (SEQ. ID No. 27), Gly Asp Lys Ile Asn Ala Met Leu Lys Gln Gln Gln Glu Val Ala Lys Leu Arg Glu Phe Ile Gln Arg Ala (SEQ. ID No. 28), Thr Phe Ser Asp Glu Glu Val Met Arg Leu Ala Glu Asn Leu Arg Lys Gly Met Pro Ieu Ala Thr Pro Val (SEQ. ID No. 29), Phe Asp Gly Ala Lys Glu Ala Glu Ile Lys Glu Leu Leu Lys Leu Gly Asp Leu Pro Thr Ser Gly Gln Ile (SEQ. ID No. 30), Arg Leu Tyr Asp Gly Arg Thr Gly Glu Gln Phe Glu Arg Pro Val Thr Val Gly Tyr Met Tyr Met Leu Lys (SEQ. ID No. 31), Arg Pro Val Thr Val Gly Tyr Met Tyr Met Leu Lys Leu Asn His Leu Val Asp Asp Lys Met His Ala Arg (SEQ. ID No. 32), Tyr Asp Leu Gly Ala Asp Val Arg Gln Lys Val Asp Leu Ser Thr Phe Ser Asp Glu Glu Val Met Arg Leu (SEQ. ID No. 33), Ala Glu Asn Leu Arg Lys Gly Met Pro Ieu Ala Thr Pro Val Phe Asp Gly Ala Lys Glu Ala Glu Ile Lys Glu Leu (SEQ. ID No. 34), Leu Lys Leu Gly Asp Leu Pro Thr Ser Gly Gln Ile Arg Leu Tyr Asp Gly Arg Thr (SEQ. ID No. 35), and Thr Gly Glu Gln Phe Glu Arg Pro Val Thr Val Gly Tyr Met Tyr Met Leu Lys Leu Asn His Leu (SEQ. ID No. 36).

81. The method of Claim 65 wherein the compound blocks the binding of the β′-subunit to the β-subunit.

82. The method of Claim 81 wherein the compound binds to β-subunit amino acid residues 1247 through 1342.

83. The method of Claim 81 wherein the compound is or binds to an amino acid sequence selected from the group consisting of peptides Ser Thr Gly Ser Tyr Ser Leu Val Thr Gln Gln Pro Leu Gly Gly Lys Ala Gln Phe Gly (SEQ. ID No. 37), Gly Gln Arg Phe Gly Glu Met Glu Val Trp Ala Leu Glu Ala Tyr Gly Ala Ala Tyr Thr Leu Gln Glu Met (SEQ. ID No. 38), Met Leu Thr Val Lys Ser Asp Asp Val Asn Gly Arg Thr Lys Met Tyr Lys Asn Ile Val Asp Gly (SEQ. ID No. 39), Asn His Gln Met Glu Pro Gly Met Pro Glu Ser Phe Asn Val Leu Leu Lys Glu Ile Arg Ser Leu Gly (SEQ. ID No. 40), Met Pro Glu Ser Phe Asn Val Leu Leu Lys Glu Ile Arg Ser Leu Gly Ile Asn Ile Glu Leu Glu Asp Glu (SEQ. ID No. 41), Thr Gln Gln Pro Leu Gly Gly Lys Ala Gln Phe Gly Gly Gln Arg Phe Gly Glu Met Glu Val (SEQ. ID No. 42), Val Trp Ala Leu Glu Ala Tyr Gly Ala Ala Tyr Thr Leu Gln Glu Met Leu Thr Val Lys Ser Asp Asp Val (SEQ. ID No. 43), and Val Asn Gly Arg Thr Lys Met Tyr Lys Asn Ile Val Asp Gly Asn His Gln Met Glu Pro Gly (SEQ. ID No. 44).

84. The method of Claim 81 wherein the compound is a derivative of an amino acid sequence selected from the group consisting of peptides Ser Thr Gly Ser Tyr Ser Leu Val Thr Gln Gln Pro Leu Gly Gly Lys Ala Gln Phe Gly (SEQ. ID No. 37), Gly Gln Arg Phe Gly Glu Met Glu Val Trp Ala Leu Glu Ala Tyr Gly Ala Ala Tyr Thr Leu Gln Glu Met (SEQ. ID No. 38), Met Leu Thr Val Lys Ser Asp Asp Val Asn Gly Arg Thr Lys Met Tyr Lys Asn Ile Val Asp Gly (SEQ. ID No. 39), Asn His Gln Met Glu Pro Gly Met Pro Glu Ser Phe Asn Val Leu Leu Lys Glu Ile Arg Ser Leu Gly (SEQ. ID No. 40), Met Pro Glu Ser Phe Asn Val Leu Leu Lys Glu Ile Arg Ser Leu Gly Ile Asn Ile Glu Leu Glu Asp Glu (SEQ. ID No. 41), Thr Gln Gln Pro Leu Gly Gly Lys Ala Gln Phe Gly Gly Gln Arg Phe Gly Glu Met Glu Val (SEQ. ID No. 42), Val Trp Ala Leu Glu Ala Tyr Gly Ala Ala Tyr Thr Leu Gln Glu Met Leu Thr Val Lys Ser Asp Asp Val (SEQ. ID No. 43), and Val Asn Gly Arg Thr Lys Met Tyr Lys Asn Ile Val Asp Gly Asn His Gln Met Glu Pro Gly (SEQ. ID No. 44).

85. A method of killing bacterial cells comprising the step of bringing bacterial cells into contact with a compound that blocks the binding of the σ-subunit of RNAP to the RNAP core.

86. The method of Claim 85 wherein the compound binds to σ70-subunit amino acid residues 361 through 390.

87. The method of Claim 85 wherein the compound is or binds to an amino acid sequence selected from the group consisting of Ile Asn Arg Arg Met Ser Ile Gly Glu Ala Lys Ala Arg Arg Ala (SEQ. ID No. 48), Ala Lys Lys Glu Met Val Glu Ala Asn Leu Arg Leu Val Ile Ser Ile (SEQ. ID No. 49), and Met Ser Ile Gly Glu Ala Lys Ala Arg Arg Ala Lys Lys Glu Met Val Glu Ala (SEQ. ID No. 50).

88. The method of Claim 85 wherein the compound is a derivative of an amino acid sequence selected from the group consisting of Ile Asn Arg Arg Met Ser Ile Gly Glu Ala Lys Ala Arg Arg Ala (SEQ. ID No. 48), Ala Lys Lys Glu Met Val Glu Ala Asn Leu Arg Leu Val Ile Ser Ile (SEQ. ID No. 49), and Met Ser Ile Gly Glu Ala Lys Ala Arg Arg Ala Lys Lys Glu Met Val Glu Ala (SEQ. ID No. 50).

89. A method of killing bacterial cells comprising the step of bringing bacterial cells into contact with a compound that blocks nucleic acid binding to the β′-subunit of RNAP.

90. The method of Claim 89 wherein the compound binds to β`-subunit amino acid residues 1 through 61 or 298 through 330.

91. The method of Claim 89 wherein the compound is or binds to an amino acid sequence selected from the group consisting of Met Lys Asp Leu Leu Lys Phe Leu Lys Ala Gln Thr Lys Thr Glu Glu Phe Asp Ala Ile Lys Ile Ala (SEQ. ID No. 54), Ile Ala Leu Ala Ser Pro Asp Met Ile Arg Ser Trp Ser Phe Gly Glu Val Lys Lys Pro Glu Thr Ile (SEQ. ID No. 55), Thr Ile Asn Tyr Arg Thr Phe Lys Pro Glu Arg Asp Gly Leu Phe Cys Ala Arg Ile (SEQ. ID No. 56), Val Lys Lys Pro Glu Thr Ile Asn Tyr Arg Thr Phe Lys Pro Glu Arg Asp Gly Leu Phe Cys (SEQ. ID No. 57), Thr Glu Glu

Phe Asp Ala Ile Lys Ile Ala Leu Ala Ser Pro Asp Met Ile Arg Ser Trp Ser Phe Gly Glu Val (SEQ. ID No. 58), Met Leu Gln Glu Ala Val Asp Ala Leu Leu Asp Asn Gly Arg Arg Gly Arg Ala (SEQ. ID No. 59), Ala Ile Thr Gly Ser Asn Lys Lys Pro Leu Lys Ser Leu Ala Asp Met (SEQ. ID No. 60), and Leu Asp Asn Gly Arg Arg Gly Arg Ala Ile Thr Gly Ser Asn Lys Lys Pro Leu Lys Ser Leu (SEQ. ID No. 61).

92. The method of Claim 89 wherein the compound is a derivative of an amino acid sequence selected from the group consisting of Met Lys Asp Leu Leu Lys Phe Leu Lys Ala Gln Thr Lys Thr Glu Glu Phe Asp Ala Ile Lys Ile Ala (SEQ. ID No. 54), Ile Ala Leu Ala Ser Pro Asp Met Ile Arg Ser Trp Ser Phe Gly Glu Val Lys Lys Pro Glu Thr Ile (SEQ. ID No. 55), Thr Ile Asn Tyr Arg Thr Phe Lys Pro Glu Arg Asp Gly Leu Phe Cys Ala Arg Ile (SEQ. ID No. 56), Val Lys Lys Pro Glu Thr Ile Asn Tyr Arg Thr Phe Lys Pro Glu Arg Asp Gly Leu Phe Cys (SEQ. ID No. 57), Thr Glu Glu Phe Asp Ala Ile Lys Ile Ala Leu Ala Ser Pro Asp Met Ile Arg Ser Trp Ser Phe Gly Glu Val (SEQ. ID No. 58), Met Leu Gln Glu Ala Val Asp Ala Leu Leu Asp Asn Gly Arg Arg Gly Arg Ala (SEQ. ID No. 59), Ala Ile Thr Gly Ser Asn Lys Lys Pro Leu Lys Ser Leu Ala Asp Met (SEQ. ID No. 60), and Leu Asp Asn Gly Arg Arg Gly Arg Ala Ile Thr Gly Ser Asn Lys Lys Pro Leu Lys Ser Leu (SEQ. ID No. 61).

93. A method of killing bacterial cells comprising the step of bringing bacterial cells into contact with a compound that blocks nucleic acid binding to the β-subunit of RNAP.

94. The method of Claim 93 wherein the compound binds to β-subunit amino acid residues 130 through 239 or 1230 through 1304.

95. The method of Claim 93 wherein the compound is or binds to an amino acid sequence selected from the group consisting of Met Thr Asp Asn Gly Thr Phe Val Ile Asn Gly Thr Glu Arg Val Ile Val Ser Gln Leu His Arg (SEQ. ID No. 66), Ser Pro Gly Val Phe Phe Asp Ser Asp Lys Gly Lys Thr His Ser Ser Gly Lys Val Leu Tyr Asn (SEQ. ID No. 67), Ala Arg Ile Ile Pro Tyr Arg Gly Ser Trp Leu Asp Phe Glu Phe Asp Pro Lys Asp Asn Leu Phe (SEQ. ID No. 68), Val Arg Ile Asp Arg Arg Arg Lys Leu Pro Ala Thr Ile Ile Leu Arg Ala Leu Asn Tyr Thr Thr (SEQ. ID No. 69), Glu

Gln Ile Leu Asp Leu Phe Phe Glu Lys Val Ile Phe Glu Ile Arg Asp Asn Lys Leu Gln Met (SEQ. ID No. 70), Ser Trp Leu Asp Phe Glu Phe Asp Pro Lys Asp Asn Leu Phe Val Arg Ile Asp Arg Arg Arg Lys Leu Pro (SEQ. ID No. 71), Val Ser Gln Leu His Arg Ser Pro Gly Val Phe Phe Asp Ser Asp Lys Gly Lys Thr His Ser (SEQ. ID No. 72), Ser Gly Lys Val Leu Tyr Asn Ala Arg Ile Ile Pro Tyr Arg Gly Ser Trp Leu (SEQ. ID No. 73), Leu Asp Phe Glu Phe Asp Pro Lys Asp Asn Leu Phe Val Arg Ile Asp Arg Arg Arg Lys Leu Pro (SEQ. ID No. 74), Ala Thr Ile Ile Leu Arg Ala Leu Asn Tyr Thr Thr Glu Gln Ile Leu Asp Leu Phe Phe Glu Lys Val (SEQ. ID No. 75), Met Tyr Met Leu Lys Leu Asn His Leu Val Asp Asp Lys Met His Ala Arg Ser Thr Gly Ser Tyr Ser Leu Val (SEQ. ID No. 76), Thr Gln Gln Pro Leu Gly Gly Lys Ala Gln Phe Gly Gly Gln Arg Phe Gly Glu Met Glu Val Trp Ala Leu Glu (SEQ. ID No. 77), Ala Tyr Gly Ala Ala Tyr Thr Leu Gln Glu Met Leu Thr Val Lys Ser Asp Asp Val Asn Gly Arg Thr Lys Met (SEQ. ID No. 78), Met His Ala Arg Ser Thr Gly Ser Tyr Ser Leu Val Thr Gln Gln Pro Leu Gly Gly Lys Ala Gln Phe Gly (SEQ. ID No. 79)and Gly Gln Arg Phe Gly Glu Met Glu Val Trp Ala Leu Glu Ala Tyr Gly Ala Ala Tyr Thr Leu Gln Glu Met Leu (SEQ. ID No. 80).

96. The method of Claim 93 wherein the compound is a derivative of an amino acid sequence selected from the group consisting of Met Thr Asp Asn Gly Thr Phe Val Ile Asn Gly Thr Glu Arg Val Ile Val Ser Gln Leu His Arg (SEQ. ID No. 66), Ser Pro Gly Val Phe Phe Asp Ser Asp Lys Gly Lys Thr His Ser Ser Gly Lys Val Leu Tyr Asn (SEQ. ID No. 67), Ala Arg Ile Ile Pro Tyr Arg Gly Ser Trp Leu Asp Phe Glu Phe Asp Pro Lys Asp Asn Leu Phe (SEQ. ID No. 68), Val Arg Ile Asp Arg Arg Arg Lys Leu Pro Ala Thr Ile Ile Leu Arg Ala Leu Asn Tyr Thr Thr (SEQ. ID No. 69), Glu Gln Ile Leu Asp Leu Phe Phe Glu Lys Val Ile Phe Glu Ile Arg Asp Asn Lys Leu Gln Met (SEQ. ID No. 70), Ser Trp Leu Asp Phe Glu Phe Asp Pro Lys Asp Asn Leu Phe Val Arg Ile Asp Arg Arg Arg Lys Leu Pro (SEQ. ID No. 71), Val Ser Gln Leu His Arg Ser Pro Gly Val Phe Phe Asp Ser Asp Lys Gly Lys Thr His Ser (SEQ. ID No. 72), Ser Gly Lys Val Leu Tyr Asn Ala Arg Ile Ile Pro Tyr Arg Gly Ser Trp Leu (SEQ. ID No. 73), Leu Asp Phe Glu Phe Asp Pro Lys Asp Asn Leu Phe Val Arg Ile Asp Arg Arg Arg Lys Leu Pro (SEQ. ID No. 74), Ala Thr Ile Ile Leu Arg Ala Leu Asn Tyr Thr Thr Glu Gln Ile Leu Asp Leu Phe Phe Glu Lys Val (SEQ. ID No. 75), Met Tyr Met

Leu Lys Leu Asn His Leu Val Asp Asp Lys Met His Ala Arg Ser Thr Gly Ser Tyr Ser Leu Val (SEQ. ID No. 76), Thr Gln Gln Pro Leu Gly Gly Lys Ala Gln Phe Gly Gly Gln Arg Phe Gly Glu Met Glu Val Trp Ala Leu Glu (SEQ. ID No. 77), Ala Tyr Gly Ala Ala Tyr Thr Leu Gln Glu Met Leu Thr Val Lys Ser Asp Asp Val Asn Gly Arg Thr Lys Met (SEQ. ID No. 78), Met His Ala Arg Ser Thr Gly Ser Tyr Ser Leu Val Thr Gln Gln Pro Leu Gly Gly Lys Ala Gln Phe Gly (SEQ. ID No. 79)and Gly Gln Arg Phe Gly Glu Met Glu Val Trp Ala Leu Glu Ala Tyr Gly Ala Ala Tyr Thr Leu Gln Glu Met Leu (SEQ. ID No. 80).

97. A method of design of an antibacterial drug comprising:

a) identifying a region of an RNAP subunit that is involved in making subunit-subunit contacts;

b) performing a fine mapping of the region;

c) designing a compound that binds to the region;

d) translating the compound to the antibacterial drug using combinatorial chemistry; and

e) testing the effect of the drug on bacterial growth.

98. The method of Claim 97 wherein the compound blocks the binding of the σ-subunit to the β′-subunit.

99. The method of Claim 97 wherein the compound binds to β'-subunit amino acid residues 60 through 135.

100. The method of Claim 98 wherein the compound is or binds to an amino acid sequence selected from the group consisting of
RIFGPVKDYECLCGKYKRLKHRGVICEKCGVEVTQTKVRRERMGHIELASPT AHIWFLKSLPSRIGLLLDMPLRDI (SEQ ID NO:4);
ARIFGPVKDHECLCGKYKRLKHRG (SEQ ID NO:5);
ICEKCGVEVTQTKVRRERMGHI (SEQ ID NO:6);

CHIWFLKSLPSRIGLLLDMPLRDIE (SEQ ID NO:7);
ECLCGKYKRLKHRGVICEKCGV (SEQ ID NO:8); and
CKVRRERMGHIELASPTAHIWFLKSL (SEQ ID NO:9).

101. The method of Claim 98 wherein the compound is a derivative of an amino acid sequence selected from the group consisting of
RIFGPVKDYECLCGKYKRLKHRGVICEKCGVEVTQTKVRRERMGHIELASPT
AHIWFLKSLPSRIGLLLDMPLRDI (SEQ ID NO:4);
ARIFGPVKDHECLCGKYKRLKHRG (SEQ ID NO:5);
ICEKCGVEVTQTKVRRERMGHI (SEQ ID NO:6);
CHIWFLKSLPSRIGLLLDMPLRDIE (SEQ ID NO:7);
ECLCGKYKRLKHRGVICEKCGV (SEQ ID NO:8); and
CKVRRERMGHIELASPTAHIWFLKSL (SEQ ID NO:9).

102. The method of Claim 97 wherein the compound blocks the binding of the α-subunit to the β′-subunit.

103. The method of Claim 102 wherein the compound binds to α-subunit amino acid residues 175 through 185 or 195 through 210.

104. The method of Claim 102 wherein the compound is or binds to an amino acid sequence selected from the group consisting of peptides Leu Val Asp Ala Cys Tyr Ser Pro Val Glu Arg Ile Ala Tyr Asn Val Glu Ala (SEQ. ID No. 21), Ala Ala Arg Val Glu Gln Arg Thr Asp Leu Asp Lys Leu Val Ile Glu Met Glu Thr Asn Gly Thr (SEQ. ID No. 22), and Val Glu Arg Ile Ala Tyr Asn Val Glu Ala Ala Arg Val Glu Gln Arg Thr Asp Leu Asp Lys Leu Val (SEQ. ID No. 23).

105. The method of Claim 102 wherein the compound is a derivative of an amino acid sequence selected from the group consisting of peptides Leu Val Asp Ala Cys Tyr Ser Pro Val Glu Arg Ile Ala Tyr Asn Val Glu Ala (SEQ. ID No. 21), Ala Ala Arg Val Glu Gln Arg Thr Asp Leu Asp Lys Leu Val Ile Glu Met Glu Thr Asn Gly Thr (SEQ. ID No. 22), and Val Glu Arg Ile Ala Tyr Asn Val Glu Ala Ala Arg Val Glu Gln Arg Thr Asp Leu Asp Lys Leu Val (SEQ. ID No. 23).

106. The method of Claim 97 wherein the compound blocks the binding of the α-subunit to the β-subunit.

107. The method of Claim 106 wherein the compound binds to α-subunit amino acid residues 30 through 55 or 61 through 76.

108. The method of Claim 106 wherein the compound is or binds to an amino acid sequence selected from the group consisting of peptides Pro Leu Glu Arg Gly Phe Gly His Thr Leu Gly Asn Ala Leu Arg Arg Ile Leu Leu Ser Met Pro Gly Cys (SEQ. ID No. 18), Cys Ala Val Thr Glu Val Glu Ile Asp Gly Val Leu His Glu Tyr Ser Thr Lys Glu Gly Val Gln Glu Asp Ile (SEQ. ID No. 19), and Asn Ala Leu Arg Arg Ile Leu Leu Ser Met Pro Gly Cys AlaVal Thr Glu Val Glu Ile Asp Gly Val Leu (SEQ. ID No. 20).

109. The method of Claim 106 wherein the compound is a derivative of an amino acid sequence selected from the group consisting of peptides Pro Leu Glu Arg Gly Phe Gly His Thr Leu Gly Asn Ala Leu Arg Arg Ile Leu Leu Ser Met Pro Gly Cys (SEQ. ID No. 18), Cys Ala Val Thr Glu Val Glu Ile Asp Gly Val Leu His Glu Tyr Ser Thr Lys Glu Gly Val Gln Glu Asp Ile (SEQ. ID No. 19), and Asn Ala Leu Arg Arg Ile Leu Leu Ser Met Pro Gly Cys AlaVal Thr Glu Val Glu Ile Asp Gly Val Leu (SEQ. ID No. 20).

110. The method of Claim 106 wherein the compound binds to β-subunit amino acid residues 907 through 1246.

111. The method of Claim 106 wherein the compound is or binds to an amino acid sequence selected from the group consisting of peptides Thr His Leu Gly Met Ala Ala Lys Gly Ile Gly Asp Lys Ile Asn Ala Met Leu Lys Gln Gln Gln Glu Val (SEQ. ID No. 26), Ala Lys Leu Arg Glu Phe Ile Gln Arg Ala Tyr Asp Leu Gly Ala Asp Val Arg Gln Lys Val Asp Leu Ser (SEQ. ID No. 27), Gly Asp Lys Ile Asn Ala Met Leu Lys Gln Gln Gln Glu Val Ala Lys Leu Arg Glu Phe Ile Gln Arg Ala (SEQ. ID No. 28), Thr Phe Ser Asp Glu Glu Val Met Arg Leu Ala Glu Asn Leu Arg Lys Gly Met Pro Ieu Ala Thr Pro Val (SEQ. ID No. 29), Phe Asp Gly Ala Lys Glu Ala Glu Ile

Lys Glu Leu Leu Lys Leu Gly Asp Leu Pro Thr Ser Gly Gln Ile (SEQ. ID No. 30), Arg Leu Tyr Asp Gly Arg Thr Gly Glu Gln Phe Glu Arg Pro Val Thr Val Gly Tyr Met Tyr Met Leu Lys (SEQ. ID No. 31), Arg Pro Val Thr Val Gly Tyr Met Tyr Met Leu Lys Leu Asn His Leu Val Asp Asp Lys Met His Ala Arg (SEQ. ID No. 32), Tyr Asp Leu Gly Ala Asp Val Arg Gln Lys Val Asp Leu Ser Thr Phe Ser Asp Glu Glu Val Met Arg Leu (SEQ. ID No. 33), Ala Glu Asn Leu Arg Lys Gly Met Pro Ieu Ala Thr Pro Val Phe Asp Gly Ala Lys Glu Ala Glu Ile Lys Glu Leu (SEQ. ID No. 34), Leu Lys Leu Gly Asp Leu Pro Thr Ser Gly Gln Ile Arg Leu Tyr Asp Gly Arg Thr (SEQ. ID No. 35), and Thr Gly Glu Gln Phe Glu Arg Pro Val Thr Val Gly Tyr Met Tyr Met Leu Lys Leu Asn His Leu (SEQ. ID No. 36).

112. The method of Claim 106 wherein the compound is a derivative of an amino acid sequence selected from the group consisting of peptides Thr His Leu Gly Met Ala Ala Lys Gly Ile Gly Asp Lys Ile Asn Ala Met Leu Lys Gln Gln Gln Glu Val (SEQ. ID No. 26), Ala Lys Leu Arg Glu Phe Ile Gln Arg Ala Tyr Asp Leu Gly Ala Asp Val Arg Gln Lys Val Asp Leu Ser (SEQ. ID No. 27), Gly Asp Lys Ile Asn Ala Met Leu Lys Gln Gln Gln Glu Val Ala Lys Leu Arg Glu Phe Ile Gln Arg Ala (SEQ. ID No. 28), Thr Phe Ser Asp Glu Glu Val Met Arg Leu Ala Glu Asn Leu Arg Lys Gly Met Pro Ieu Ala Thr Pro Val (SEQ. ID No. 29), Phe Asp Gly Ala Lys Glu Ala Glu Ile Lys Glu Leu Leu Lys Leu Gly Asp Leu Pro Thr Ser Gly Gln Ile (SEQ. ID No. 30), Arg Leu Tyr Asp Gly Arg Thr Gly Glu Gln Phe Glu Arg Pro Val Thr Val Gly Tyr Met Tyr Met Leu Lys (SEQ. ID No. 31), Arg Pro Val Thr Val Gly Tyr Met Tyr Met Leu Lys Leu Asn His Leu Val Asp Asp Lys Met His Ala Arg (SEQ. ID No. 32), Tyr Asp Leu Gly Ala Asp Val Arg Gln Lys Val Asp Leu Ser Thr Phe Ser Asp Glu Glu Val Met Arg Leu (SEQ. ID No. 33), Ala Glu Asn Leu Arg Lys Gly Met Pro Ieu Ala Thr Pro Val Phe Asp Gly Ala Lys Glu Ala Glu Ile Lys Glu Leu (SEQ. ID No. 34), Leu Lys Leu Gly Asp Leu Pro Thr Ser Gly Gln Ile Arg Leu Tyr Asp Gly Arg Thr (SEQ. ID No. 35), and Thr Gly Glu Gln Phe Glu Arg Pro Val Thr Val Gly Tyr Met Tyr Met Leu Lys Leu Asn His Leu (SEQ. ID No. 36).

113. The method of Claim 97 wherein the compound blocks the binding of the β′-subunit to the β-subunit.

114. The method of Claim 113 wherein the compound binds to β amino acid residues 1247 through 1342.

115. The method of Claim 113 wherein the compound is or binds to an amino acid sequence selected from the group consisting of peptides Ser Thr Gly Ser Tyr Ser Leu Val Thr Gln Gln Pro Leu Gly Gly Lys Ala Gln Phe Gly (SEQ. ID No. 37), Gly Gln Arg Phe Gly Glu Met Glu Val Trp Ala Leu Glu Ala Tyr Gly Ala Ala Tyr Thr Leu Gln Glu Met (SEQ. ID No. 38), Met Leu Thr Val Lys Ser Asp Asp Val Asn Gly Arg Thr Lys Met Tyr Lys Asn Ile Val Asp Gly (SEQ. ID No. 39), Asn His Gln Met Glu Pro Gly Met Pro Glu Ser Phe Asn Val Leu Leu Lys Glu Ile Arg Ser Leu Gly (SEQ. ID No. 40), Met Pro Glu Ser Phe Asn Val Leu Leu Lys Glu Ile Arg Ser Leu Gly Ile Asn Ile Glu Leu Glu Asp Glu (SEQ. ID No. 41), Thr Gln Gln Pro Leu Gly Gly Lys Ala Gln Phe Gly Gly Gln Arg Phe Gly Glu Met Glu Val (SEQ. ID No. 42), Val Trp Ala Leu Glu Ala Tyr Gly Ala Ala Tyr Thr Leu Gln Glu Met Leu Thr Val Lys Ser Asp Asp Val (SEQ. ID No. 43), and Val Asn Gly Arg Thr Lys Met Tyr Lys Asn Ile Val Asp Gly Asn His Gln Met Glu Pro Gly (SEQ. ID No. 44).

116. The method of Claim 113 wherein the compound is a derivative of an amino acid sequence selected from the group consisting of peptides Ser Thr Gly Ser Tyr Ser Leu Val Thr Gln Gln Pro Leu Gly Gly Lys Ala Gln Phe Gly (SEQ. ID No. 37), Gly Gln Arg Phe Gly Glu Met Glu Val Trp Ala Leu Glu Ala Tyr Gly Ala Ala Tyr Thr Leu Gln Glu Met (SEQ. ID No. 38), Met Leu Thr Val Lys Ser Asp Asp Val Asn Gly Arg Thr Lys Met Tyr Lys Asn Ile Val Asp Gly (SEQ. ID No. 39), Asn His Gln Met Glu Pro Gly Met Pro Glu Ser Phe Asn Val Leu Leu Lys Glu Ile Arg Ser Leu Gly (SEQ. ID No. 40), Met Pro Glu Ser Phe Asn Val Leu Leu Lys Glu Ile Arg Ser Leu Gly Ile Asn Ile Glu Leu Glu Asp Glu (SEQ. ID No. 41), Thr Gln Gln Pro Leu Gly Gly Lys Ala Gln Phe Gly Gly Gln Arg Phe Gly Glu Met Glu Val (SEQ. ID No. 42), Val Trp Ala Leu Glu Ala Tyr Gly Ala Ala Tyr Thr Leu Gln Glu Met Leu Thr Val Lys Ser Asp Asp Val (SEQ. ID No. 43), and Val Asn Gly Arg Thr Lys Met Tyr Lys Asn Ile Val Asp Gly Asn His Gln Met Glu Pro Gly (SEQ. ID No. 44).

117. A method of design of an antibacterial drug comprising:

a) identifying a region of an RNAP subunit that is involved in the binding of the σ-subunit of RNAP to the RNAP core;

b) performing a fine mapping of the region;

c) designing a compound that binds to the region;

d) translating the compound to the antibacterial drug using combinatorial chemistry; and

e) testing the effect of the drug on bacterial growth.

118. The method of Claim 117 wherein the compound binds to σ70-subunit amino acid residues 361 through 390.

119. The method of Claim 117 wherein the compound is or binds to an amino acid sequence selected from the group consisting of Ile Asn Arg Arg Met Ser Ile Gly Glu Ala Lys Ala Arg Arg Ala (SEQ. ID No. 48), Ala Lys Lys Glu Met Val Glu Ala Asn Leu Arg Leu Val Ile Ser Ile (SEQ. ID No. 49), and Met Ser Ile Gly Glu Ala Lys Ala Arg Arg Ala Lys Lys Glu Met Val Glu Ala (SEQ. ID No. 50).

120. The method of Claim 117 wherein the compound is a derivative of an amino acid sequence selected from the group consisting of Ile Asn Arg Arg Met Ser Ile Gly Glu Ala Lys Ala Arg Arg Ala (SEQ. ID No. 48), Ala Lys Lys Glu Met Val Glu Ala Asn Leu Arg Leu Val Ile Ser Ile (SEQ. ID No. 49), and Met Ser Ile Gly Glu Ala Lys Ala Arg Arg Ala Lys Lys Glu Met Val Glu Ala (SEQ. ID No. 50).

121. A method of design of an antibacterial drug comprising:

a) identifying a region of an RNAP subunit that is involved in the nucleic acid binding to the β′-subunit of RNAP;

b) performing a fine mapping of the region;

c) designing a compound that binds to the region;

d) translating the compound to the antibacterial drug using combinatorial chemistry; and

e) testing the effect of the drug on bacterial growth.

122. The method of Claim 121 wherein the compound binds to β'-subunit amino acid residues 1 through 61 or 298 through 330.

123. The method of Claim 121 wherein the compound is or binds to an amino acid sequence selected from the group consisting of Met Lys Asp Leu Leu Lys Phe Leu Lys Ala Gln Thr Lys Thr Glu Glu Phe Asp Ala Ile Lys Ile Ala (SEQ. ID No. 54), Ile Ala Leu Ala Ser Pro Asp Met Ile Arg Ser Trp Ser Phe Gly Glu Val Lys Lys Pro Glu Thr Ile (SEQ. ID No. 55), Thr Ile Asn Tyr Arg Thr Phe Lys Pro Glu Arg Asp Gly Leu Phe Cys Ala Arg Ile (SEQ. ID No. 56), Val Lys Lys Pro Glu Thr Ile Asn Tyr Arg Thr Phe Lys Pro Glu Arg Asp Gly Leu Phe Cys (SEQ. ID No. 57), Thr Glu Glu Phe Asp Ala Ile Lys Ile Ala Leu Ala Ser Pro Asp Met Ile Arg Ser Trp Ser Phe Gly Glu Val (SEQ. ID No. 58), Met Leu Gln Glu Ala Val Asp Ala Leu Leu Asp Asn Gly Arg Arg Gly Arg Ala (SEQ. ID No. 59), Ala Ile Thr Gly Ser Asn Lys Lys Pro Leu Lys Ser Leu Ala Asp Met (SEQ. ID No. 60), and Leu Asp Asn Gly Arg Arg Gly Arg Ala Ile Thr Gly Ser Asn Lys Lys Pro Leu Lys Ser Leu (SEQ. ID No. 61).

124. The method of Claim 121 wherein the compound is a derivative of an amino acid sequence selected from the group consisting of Met Lys Asp Leu Leu Lys Phe Leu Lys Ala Gln Thr Lys Thr Glu Glu Phe Asp Ala Ile Lys Ile Ala (SEQ. ID No. 54), Ile Ala Leu Ala Ser Pro Asp Met Ile Arg Ser Trp Ser Phe Gly Glu Val Lys Lys Pro Glu Thr Ile (SEQ. ID No. 55), Thr Ile Asn Tyr Arg Thr Phe Lys Pro Glu Arg Asp Gly Leu Phe Cys Ala Arg Ile (SEQ. ID No. 56), Val Lys Lys Pro Glu Thr Ile Asn Tyr Arg Thr Phe Lys Pro Glu Arg Asp Gly Leu Phe Cys (SEQ. ID No. 57), Thr Glu Glu Phe Asp Ala Ile Lys Ile Ala Leu Ala Ser Pro Asp Met Ile Arg Ser Trp Ser Phe Gly Glu Val (SEQ. ID No. 58), Met Leu Gln Glu Ala Val Asp Ala Leu Leu Asp Asn Gly Arg Arg Gly Arg Ala (SEQ. ID No. 59), Ala Ile Thr Gly Ser Asn Lys Lys Pro Leu Lys Ser Leu Ala Asp Met (SEQ. ID No. 60), and Leu Asp Asn Gly

Arg Arg Gly Arg Ala Ile Thr Gly Ser Asn Lys Lys Pro Leu Lys Ser Leu (SEQ. ID No. 61).

125. A method of design of an antibacterial drug comprising:

a) identifying a region of an RNAP subunit that is involved in nucleic acid binding to the β-subunit of RNAP;

b) performing a fine mapping of the region;

c) designing a compound that binds to the region;

d) translating the compound to the antibacterial drug using combinatorial chemistry; and

e) testing the effect of the drug on bacterial growth.

126. The method of Claim 125 wherein the compound binds to β amino acid residues 130 through 239 or 1230 through 1304.

127. The method of Claim 125 wherein the compound is or binds to an amino acid sequence selected from the group consisting of Met Thr Asp Asn Gly Thr Phe Val Ile Asn Gly Thr Glu Arg Val Ile Val Ser Gln Leu His Arg (SEQ. ID No. 66), Ser Pro Gly Val Phe Phe Asp Ser Asp Lys Gly Lys Thr His Ser Ser Gly Lys Val Leu Tyr Asn (SEQ. ID No. 67), Ala Arg Ile Ile Pro Tyr Arg Gly Ser Trp Leu Asp Phe Glu Phe Asp Pro Lys Asp Asn Leu Phe (SEQ. ID No. 68), Val Arg Ile Asp Arg Arg Arg Lys Leu Pro Ala Thr Ile Ile Leu Arg Ala Leu Asn Tyr Thr Thr (SEQ. ID No. 69), Glu Gln Ile Leu Asp Leu Phe Phe Glu Lys Val Ile Phe Glu Ile Arg Asp Asn Lys Leu Gln Met (SEQ. ID No. 70), Ser Trp Leu Asp Phe Glu Phe Asp Pro Lys Asp Asn Leu Phe Val Arg Ile Asp Arg Arg Arg Lys Leu Pro (SEQ. ID No. 71), Val Ser Gln Leu His Arg Ser Pro Gly Val Phe Phe Asp Ser Asp Lys Gly Lys Thr His Ser (SEQ. ID No. 72), Ser Gly Lys Val Leu Tyr Asn Ala Arg Ile Ile Pro Tyr Arg Gly Ser Trp Leu (SEQ. ID No. 73), Leu Asp Phe Glu Phe Asp Pro Lys Asp Asn Leu Phe Val Arg Ile Asp Arg Arg Arg Lys Leu Pro (SEQ. ID No. 74), Ala Thr Ile Ile Leu Arg Ala Leu Asn Tyr Thr Thr Glu Gln Ile Leu Asp Leu Phe Phe Glu Lys Val (SEQ. ID No. 75), Met Tyr Met

Leu Lys Leu Asn His Leu Val Asp Asp Lys Met His Ala Arg Ser Thr Gly Ser Tyr Ser Leu Val (SEQ. ID No. 76), Thr Gln Gln Pro Leu Gly Gly Lys Ala Gln Phe Gly Gly Gln Arg Phe Gly Glu Met Glu Val Trp Ala Leu Glu (SEQ. ID No. 77), Ala Tyr Gly Ala Ala Tyr Thr Leu Gln Glu Met Leu Thr Val Lys Ser Asp Asp Val Asn Gly Arg Thr Lys Met (SEQ. ID No. 78), Met His Ala Arg Ser Thr Gly Ser Tyr Ser Leu Val Thr Gln Gln Pro Leu Gly Gly Lys Ala Gln Phe Gly (SEQ. ID No. 79)and Gly Gln Arg Phe Gly Glu Met Glu Val Trp Ala Leu Glu Ala Tyr Gly Ala Ala Tyr Thr Leu Gln Glu Met Leu (SEQ. ID No. 80).

128. The method of Claim 125 wherein the compound is a derivative of an amino acid sequence selected from the group consisting of Met Thr Asp Asn Gly Thr Phe Val Ile Asn Gly Thr Glu Arg Val Ile Val Ser Gln Leu His Arg (SEQ. ID No. 66), Ser Pro Gly Val Phe Phe Asp Ser Asp Lys Gly Lys Thr His Ser Ser Gly Lys Val Leu Tyr Asn (SEQ. ID No. 67), Ala Arg Ile Ile Pro Tyr Arg Gly Ser Trp Leu Asp Phe Glu Phe Asp Pro Lys Asp Asn Leu Phe (SEQ. ID No. 68), Val Arg Ile Asp Arg Arg Arg Lys Leu Pro Ala Thr Ile Ile Leu Arg Ala Leu Asn Tyr Thr Thr (SEQ. ID No. 69), Glu Gln Ile Leu Asp Leu Phe Phe Glu Lys Val Ile Phe Glu Ile Arg Asp Asn Lys Leu Gln Met (SEQ. ID No. 70), Ser Trp Leu Asp Phe Glu Phe Asp Pro Lys Asp Asn Leu Phe Val Arg Ile Asp Arg Arg Arg Lys Leu Pro (SEQ. ID No. 71), Val Ser Gln Leu His Arg Ser Pro Gly Val Phe Phe Asp Ser Asp Lys Gly Lys Thr His Ser (SEQ. ID No. 72), Ser Gly Lys Val Leu Tyr Asn Ala Arg Ile Ile Pro Tyr Arg Gly Ser Trp Leu (SEQ. ID No. 73), Leu Asp Phe Glu Phe Asp Pro Lys Asp Asn Leu Phe Val Arg Ile Asp Arg Arg Arg Lys Leu Pro (SEQ. ID No. 74), Ala Thr Ile Ile Leu Arg Ala Leu Asn Tyr Thr Thr Glu Gln Ile Leu Asp Leu Phe Phe Glu Lys Val (SEQ. ID No. 75), Met Tyr Met Leu Lys Leu Asn His Leu Val Asp Asp Lys Met His Ala Arg Ser Thr Gly Ser Tyr Ser Leu Val (SEQ. ID No. 76), Thr Gln Gln Pro Leu Gly Gly Lys Ala Gln Phe Gly Gly Gln Arg Phe Gly Glu Met Glu Val Trp Ala Leu Glu (SEQ. ID No. 77), Ala Tyr Gly Ala Ala Tyr Thr Leu Gln Glu Met Leu Thr Val Lys Ser Asp Asp Val Asn Gly Arg Thr Lys Met (SEQ. ID No. 78), Met His Ala Arg Ser Thr Gly Ser Tyr Ser Leu Val Thr Gln Gln Pro Leu Gly Gly Lys Ala Gln Phe Gly (SEQ. ID No. 79)and Gly Gln Arg Phe Gly Glu Met Glu Val Trp Ala Leu Glu Ala Tyr Gly Ala Ala Tyr Thr Leu Gln Glu Met Leu (SEQ. ID No. 80).

129. An antibacterial drug comprising a compound that inhibits the binding of at least one protein subunit of RNAP to a second protein subunit of RNAP.

130. The drug of Claim 129 wherein the compound blocks the binding of the σ-subunit to the β′-subunit.

131. The drug of Claim 130 wherein the compound binds to β'-subunit amino acid residues 60 through 135.

132. The drug of Claim 130 wherein the compound is or binds to an amino acid sequence selected from the group consisting of
RIFGPVKDYECLCGKYKRLKHRGVICEKCGVEVTQTKVRRERMGHIELASPT
AHIWFLKSLPSRIGLLLDMPLRDI (SEQ ID NO:4);
ARIFGPVKDHECLCGKYKRLKHRG (SEQ ID NO:5);
ICEKCGVEVTQTKVRRERMGHI (SEQ ID NO:6);
CHIWFLKSLPSRIGLLLDMPLRDIE (SEQ ID NO:7);
ECLCGKYKRLKHRGVICEKCGV (SEQ ID NO:8); and
CKVRRERMGHIELASPTAHIWFLKSL (SEQ ID NO:9).

133. The drug of Claim 130 wherein the compound is a derivative of an amino acid sequence selected from the group consisting of
RIFGPVKDYECLCGKYKRLKHRGVICEKCGVEVTQTKVRRERMGHIELASPT
AHIWFLKSLPSRIGLLLDMPLRDI (SEQ ID NO:4);
ARIFGPVKDHECLCGKYKRLKHRG (SEQ ID NO:5);
ICEKCGVEVTQTKVRRERMGHI (SEQ ID NO:6);
CHIWFLKSLPSRIGLLLDMPLRDIE (SEQ ID NO:7);
ECLCGKYKRLKHRGVICEKCGV (SEQ ID NO:8); and
CKVRRERMGHIELASPTAHIWFLKSL (SEQ ID NO:9).

134. The drug of Claim 129 wherein the compound blocks the binding of the α-subunit to the β′-subunit.

135. The drug of Claim 134 wherein the compound binds to α amino acid residues 175 through 185 or 195 through 210.

136. The drug of Claim 134 wherein the compound is or binds to an amino acid sequence selected from the group consisting of peptides Leu Val Asp Ala Cys Tyr Ser Pro Val Glu Arg Ile Ala Tyr Asn Val Glu Ala (SEQ. ID No. 21), Ala Ala Arg Val Glu Gln Arg Thr Asp Leu Asp Lys Leu Val Ile Glu Met Glu Thr Asn Gly Thr (SEQ. ID No. 22), and Val Glu Arg Ile Ala Tyr Asn Val Glu Ala Ala Arg Val Glu Gln Arg Thr Asp Leu Asp Lys Leu Val (SEQ. ID No. 23).

137. The drug of Claim 134 wherein the compound is a derivative of an amino acid sequence selected from the group consisting of peptides Leu Val Asp Ala Cys Tyr Ser Pro Val Glu Arg Ile Ala Tyr Asn Val Glu Ala (SEQ. ID No. 21), Ala Ala Arg Val Glu Gln Arg Thr Asp Leu Asp Lys Leu Val Ile Glu Met Glu Thr Asn Gly Thr (SEQ. ID No. 22), and Val Glu Arg Ile Ala Tyr Asn Val Glu Ala Ala Arg Val Glu Gln Arg Thr Asp Leu Asp Lys Leu Val (SEQ. ID No. 23).

138. The drug of Claim 129 wherein the compound blocks the binding of the α-subunit to the β-subunit.

139. The drug of Claim 138 wherein the compound binds to α amino acid residues 30 through 55 or 61 through 76.

140. The drug of Claim 138 wherein the compound is or binds to an amino acid sequence selected from the group consisting of peptides Pro Leu Glu Arg Gly Phe Gly His Thr Leu Gly Asn Ala Leu Arg Arg Ile Leu Leu Ser Met Pro Gly Cys (SEQ. ID No. 18), Cys Ala Val Thr Glu Val Glu Ile Asp Gly Val Leu His Glu Tyr Ser Thr Lys Glu Gly Val Gln Glu Asp Ile (SEQ. ID No. 19), and Asn Ala Leu Arg Arg Ile Leu Leu Ser Met Pro Gly Cys AlaVal Thr Glu Val Glu Ile Asp Gly Val Leu (SEQ. ID No. 20).

141. The drug of Claim 138 wherein the compound is a derivative of an amino acid sequence selected from the group consisting of peptides Pro Leu Glu

Arg Gly Phe Gly His Thr Leu Gly Asn Ala Leu Arg Arg Ile Leu Leu Ser Met Pro Gly Cys (SEQ. ID No. 18), Cys Ala Val Thr Glu Val Glu Ile Asp Gly Val Leu His Glu Tyr Ser Thr Lys Glu Gly Val Gln Glu Asp Ile (SEQ. ID No. 19), and Asn Ala Leu Arg Arg Ile Leu Leu Ser Met Pro Gly Cys AlaVal Thr Glu Val Glu Ile Asp Gly Val Leu (SEQ. ID No. 20).

142. The drug of Claim 138 wherein the compound binds to β-subunit amino acid residues 907 through 1246.

143. The drug of Claim 138 wherein the compound is or binds to an amino acid sequence selected from the group consisting of peptides Thr His Leu Gly Met Ala Ala Lys Gly Ile Gly Asp Lys Ile Asn Ala Met Leu Lys Gln Gln Gln Glu Val (SEQ. ID No. 26), Ala Lys Leu Arg Glu Phe Ile Gln Arg Ala Tyr Asp Leu Gly Ala Asp Val Arg Gln Lys Val Asp Leu Ser (SEQ. ID No. 27), Gly Asp Lys Ile Asn Ala Met Leu Lys Gln Gln Gln Glu Val Ala Lys Leu Arg Glu Phe Ile Gln Arg Ala (SEQ. ID No. 28), Thr Phe Ser Asp Glu Glu Val Met Arg Leu Ala Glu Asn Leu Arg Lys Gly Met Pro Ieu Ala Thr Pro Val (SEQ. ID No. 29), Phe Asp Gly Ala Lys Glu Ala Glu Ile Lys Glu Leu Leu Lys Leu Gly Asp Leu Pro Thr Ser Gly Gln Ile (SEQ. ID No. 30), Arg Leu Tyr Asp Gly Arg Thr Gly Glu Gln Phe Glu Arg Pro Val Thr Val Gly Tyr Met Tyr Met Leu Lys (SEQ. ID No. 31), Arg Pro Val Thr Val Gly Tyr Met Tyr Met Leu Lys Leu Asn His Leu Val Asp Asp Lys Met His Ala Arg (SEQ. ID No. 32), Tyr Asp Leu Gly Ala Asp Val Arg Gln Lys Val Asp Leu Ser Thr Phe Ser Asp Glu Glu Val Met Arg Leu (SEQ. ID No. 33), Ala Glu Asn Leu Arg Lys Gly Met Pro Ieu Ala Thr Pro Val Phe Asp Gly Ala Lys Glu Ala Glu Ile Lys Glu Leu (SEQ. ID No. 34), Leu Lys Leu Gly Asp Leu Pro Thr Ser Gly Gln Ile Arg Leu Tyr Asp Gly Arg Thr (SEQ. ID No. 35), and Thr Gly Glu Gln Phe Glu Arg Pro Val Thr Val Gly Tyr Met Tyr Met Leu Lys Leu Asn His Leu (SEQ. ID No. 36).

144. The drug of Claim 138 wherein the compound is a derivative of an amino acid sequence selected from the group consisting of peptides Thr His Leu Gly Met Ala Ala Lys Gly Ile Gly Asp Lys Ile Asn Ala Met Leu Lys Gln Gln Gln Glu Val (SEQ. ID No. 26), Ala Lys Leu Arg Glu Phe Ile Gln Arg Ala Tyr Asp Leu Gly Ala Asp Val Arg Gln Lys Val Asp Leu Ser (SEQ. ID No. 27), Gly Asp Lys Ile Asn

Ala Met Leu Lys Gln Gln Gln Glu Val Ala Lys Leu Arg Glu Phe Ile Gln Arg Ala (SEQ. ID No. 28), Thr Phe Ser Asp Glu Glu Val Met Arg Leu Ala Glu Asn Leu Arg Lys Gly Met Pro Ieu Ala Thr Pro Val (SEQ. ID No. 29), Phe Asp Gly Ala Lys Glu Ala Glu Ile Lys Glu Leu Leu Lys Leu Gly Asp Leu Pro Thr Ser Gly Gln Ile (SEQ. ID No. 30), Arg Leu Tyr Asp Gly Arg Thr Gly Glu Gln Phe Glu Arg Pro Val Thr Val Gly Tyr Met Tyr Met Leu Lys (SEQ. ID No. 31), Arg Pro Val Thr Val Gly Tyr Met Tyr Met Leu Lys Leu Asn His Leu Val Asp Asp Lys Met His Ala Arg (SEQ. ID No. 32), Tyr Asp Leu Gly Ala Asp Val Arg Gln Lys Val Asp Leu Ser Thr Phe Ser Asp Glu Glu Val Met Arg Leu (SEQ. ID No. 33), Ala Glu Asn Leu Arg Lys Gly Met Pro Ieu Ala Thr Pro Val Phe Asp Gly Ala Lys Glu Ala Glu Ile Lys Glu Leu (SEQ. ID No. 34), Leu Lys Leu Gly Asp Leu Pro Thr Ser Gly Gln Ile Arg Leu Tyr Asp Gly Arg Thr (SEQ. ID No. 35), and Thr Gly Glu Gln Phe Glu Arg Pro Val Thr Val Gly Tyr Met Tyr Met Leu Lys Leu Asn His Leu (SEQ. ID No. 36).

145. The drug of Claim 129 wherein the compound blocks the binding of the β′-subunit to the β-subunit.

146. The drug of Claim 145 wherein the compound binds to β-subunit amino acid residues 1247 through 1342.

147. The drug of Claim 145 wherein the compound is or binds to an amino acid sequence selected from the group consisting of peptides Ser Thr Gly Ser Tyr Ser Leu Val Thr Gln Gln Pro Leu Gly Gly Lys Ala Gln Phe Gly (SEQ. ID No. 37), Gly Gln Arg Phe Gly Glu Met Glu Val Trp Ala Leu Glu Ala Tyr Gly Ala Ala Tyr Thr Leu Gln Glu Met (SEQ. ID No. 38), Met Leu Thr Val Lys Ser Asp Asp Val Asn Gly Arg Thr Lys Met Tyr Lys Asn Ile Val Asp Gly (SEQ. ID No. 39), Asn His Gln Met Glu Pro Gly Met Pro Glu Ser Phe Asn Val Leu Leu Lys Glu Ile Arg Ser Leu Gly (SEQ. ID No. 40), Met Pro Glu Ser Phe Asn Val Leu Leu Lys Glu Ile Arg Ser Leu Gly Ile Asn Ile Glu Leu Glu Asp Glu (SEQ. ID No. 41), Thr Gln Gln Pro Leu Gly Gly Lys Ala Gln Phe Gly Gly Gln Arg Phe Gly Glu Met Glu Val (SEQ. ID No. 42), Val Trp Ala Leu Glu Ala Tyr Gly Ala Ala Tyr Thr Leu Gln Glu Met Leu Thr Val Lys Ser Asp Asp Val (SEQ. ID No. 43), and Val Asn Gly Arg Thr Lys Met Tyr Lys Asn Ile Val Asp Gly Asn His Gln Met Glu Pro Gly (SEQ. ID No. 44).

148. The drug of Claim 145 wherein the compound is a derivative of an amino acid sequence selected from the group consisting of peptides Ser Thr Gly Ser Tyr Ser Leu Val Thr Gln Gln Pro Leu Gly Gly Lys Ala Gln Phe Gly (SEQ. ID No. 37), Gly Gln Arg Phe Gly Glu Met Glu Val Trp Ala Leu Glu Ala Tyr Gly Ala Ala Tyr Thr Leu Gln Glu Met (SEQ. ID No. 38), Met Leu Thr Val Lys Ser Asp Asp Val Asn Gly Arg Thr Lys Met Tyr Lys Asn Ile Val Asp Gly (SEQ. ID No. 39), Asn His Gln Met Glu Pro Gly Met Pro Glu Ser Phe Asn Val Leu Leu Lys Glu Ile Arg Ser Leu Gly (SEQ. ID No. 40), Met Pro Glu Ser Phe Asn Val Leu Leu Lys Glu Ile Arg Ser Leu Gly Ile Asn Ile Glu Leu Glu Asp Glu (SEQ. ID No. 41), Thr Gln Gln Pro Leu Gly Gly Lys Ala Gln Phe Gly Gly Gln Arg Phe Gly Glu Met Glu Val (SEQ. ID No. 42), Val Trp Ala Leu Glu Ala Tyr Gly Ala Ala Tyr Thr Leu Gln Glu Met Leu Thr Val Lys Ser Asp Asp Val (SEQ. ID No. 43), and Val Asn Gly Arg Thr Lys Met Tyr Lys Asn Ile Val Asp Gly Asn His Gln Met Glu Pro Gly (SEQ. ID No. 44).

149. A bacterial drug comprising a compound that blocks the binding of the σ-subunit of RNAP to the RNAP core.

150. The drug of Claim 149 wherein the compound binds to σ70-subunit amino acid residues 361 through 390.

151. The drug of Claim 149 wherein the compound is or binds to an amino acid sequence selected from the group consisting of Ile Asn Arg Arg Met Ser Ile Gly Glu Ala Lys Ala Arg Arg Ala (SEQ. ID No. 48), Ala Lys Lys Glu Met Val Glu Ala Asn Leu Arg Leu Val Ile Ser Ile (SEQ. ID No. 49), and Met Ser Ile Gly Glu Ala Lys Ala Arg Arg Ala Lys Lys Glu Met Val Glu Ala (SEQ. ID No. 50).

152. The drug of Claim 149 wherein the compound is a derivative of an amino acid sequence selected from the group consisting of Ile Asn Arg Arg Met Ser Ile Gly Glu Ala Lys Ala Arg Arg Ala (SEQ. ID No. 48), Ala Lys Lys Glu Met Val Glu Ala Asn Leu Arg Leu Val Ile Ser Ile (SEQ. ID No. 49), and Met Ser Ile Gly Glu Ala Lys Ala Arg Arg Ala Lys Lys Glu Met Val Glu Ala (SEQ. ID No. 50).

153. A bacterial drug comprising a compound that blocks nucleic acid binding to the β′-subunit of RNAP.

154. The drug of Claim 153 wherein the compound binds to β`-subunit amino acid residues 1 through 61 or 298 through 330.

155. The drug of Claim 153 wherein the compound is or binds to an amino acid sequence selected from the group consisting of Met Lys Asp Leu Leu Lys Phe Leu Lys Ala Gln Thr Lys Thr Glu Glu Phe Asp Ala Ile Lys Ile Ala (SEQ. ID No. 54), Ile Ala Leu Ala Ser Pro Asp Met Ile Arg Ser Trp Ser Phe Gly Glu Val Lys Lys Pro Glu Thr Ile (SEQ. ID No. 55), Thr Ile Asn Tyr Arg Thr Phe Lys Pro Glu Arg Asp Gly Leu Phe Cys Ala Arg Ile (SEQ. ID No. 56), Val Lys Lys Pro Glu Thr Ile Asn Tyr Arg Thr Phe Lys Pro Glu Arg Asp Gly Leu Phe Cys (SEQ. ID No. 57), Thr Glu Glu Phe Asp Ala Ile Lys Ile Ala Leu Ala Ser Pro Asp Met Ile Arg Ser Trp Ser Phe Gly Glu Val (SEQ. ID No. 58), Met Leu Gln Glu Ala Val Asp Ala Leu Leu Asp Asn Gly Arg Arg Gly Arg Ala (SEQ. ID No. 59), Ala Ile Thr Gly Ser Asn Lys Lys Pro Leu Lys Ser Leu Ala Asp Met (SEQ. ID No. 60), and Leu Asp Asn Gly Arg Arg Gly Arg Ala Ile Thr Gly Ser Asn Lys Lys Pro Leu Lys Ser Leu (SEQ. ID No. 61).

156. The drug of Claim 153 wherein the compound is a derivative of an amino acid sequence selected from the group consisting of Met Lys Asp Leu Leu Lys Phe Leu Lys Ala Gln Thr Lys Thr Glu Glu Phe Asp Ala Ile Lys Ile Ala (SEQ. ID No. 54), Ile Ala Leu Ala Ser Pro Asp Met Ile Arg Ser Trp Ser Phe Gly Glu Val Lys Lys Pro Glu Thr Ile (SEQ. ID No. 55), Thr Ile Asn Tyr Arg Thr Phe Lys Pro Glu Arg Asp Gly Leu Phe Cys Ala Arg Ile (SEQ. ID No. 56), Val Lys Lys Pro Glu Thr Ile Asn Tyr Arg Thr Phe Lys Pro Glu Arg Asp Gly Leu Phe Cys (SEQ. ID No. 57), Thr Glu Glu Phe Asp Ala Ile Lys Ile Ala Leu Ala Ser Pro Asp Met Ile Arg Ser Trp Ser Phe Gly Glu Val (SEQ. ID No. 58), Met Leu Gln Glu Ala Val Asp Ala Leu Leu Asp Asn Gly Arg Arg Gly Arg Ala (SEQ. ID No. 59), Ala Ile Thr Gly Ser Asn Lys Lys Pro Leu Lys Ser Leu Ala Asp Met (SEQ. ID No. 60), and Leu Asp Asn Gly Arg Arg Gly Arg Ala Ile Thr Gly Ser Asn Lys Lys Pro Leu Lys Ser Leu (SEQ. ID No. 61).

157. A bacterial drug comprising a compound that blocks nucleic acid binding to the β-subunit of RNAP.

158. The drug of Claim 157 wherein the compound binds to β-subunit amino acid residues 130 through 239 or 1230 through 1304.

159. The drug of Claim 157 wherein the compound is or binds to an amino acid sequence selected from the group consisting of Met Thr Asp Asn Gly Thr Phe Val Ile Asn Gly Thr Glu Arg Val Ile Val Ser Gln Leu His Arg (SEQ. ID No. 66), Ser Pro Gly Val Phe Phe Asp Ser Asp Lys Gly Lys Thr His Ser Ser Gly Lys Val Leu Tyr Asn (SEQ. ID No. 67), Ala Arg Ile Ile Pro Tyr Arg Gly Ser Trp Leu Asp Phe Glu Phe Asp Pro Lys Asp Asn Leu Phe (SEQ. ID No. 68), Val Arg Ile Asp Arg Arg Arg Lys Leu Pro Ala Thr Ile Ile Leu Arg Ala Leu Asn Tyr Thr Thr (SEQ. ID No. 69), Glu Gln Ile Leu Asp Leu Phe Phe Glu Lys Val Ile Phe Glu Ile Arg Asp Asn Lys Leu Gln Met (SEQ. ID No. 70), Ser Trp Leu Asp Phe Glu Phe Asp Pro Lys Asp Asn Leu Phe Val Arg Ile Asp Arg Arg Arg Lys Leu Pro (SEQ. ID No. 71), Val Ser Gln Leu His Arg Ser Pro Gly Val Phe Phe Asp Ser Asp Lys Gly Lys Thr His Ser (SEQ. ID No. 72), Ser Gly Lys Val Leu Tyr Asn Ala Arg Ile Ile Pro Tyr Arg Gly Ser Trp Leu (SEQ. ID No. 73), Leu Asp Phe Glu Phe Asp Pro Lys Asp Asn Leu Phe Val Arg Ile Asp Arg Arg Arg Lys Leu Pro (SEQ. ID No. 74), Ala Thr Ile Ile Leu Arg Ala Leu Asn Tyr Thr Thr Glu Gln Ile Leu Asp Leu Phe Phe Glu Lys Val (SEQ. ID No. 75), Met Tyr Met Leu Lys Leu Asn His Leu Val Asp Asp Lys Met His Ala Arg Ser Thr Gly Ser Tyr Ser Leu Val (SEQ. ID No. 76), Thr Gln Gln Pro Leu Gly Gly Lys Ala Gln Phe Gly Gly Gln Arg Phe Gly Glu Met Glu Val Trp Ala Leu Glu (SEQ. ID No. 77), Ala Tyr Gly Ala Ala Tyr Thr Leu Gln Glu Met Leu Thr Val Lys Ser Asp Asp Val Asn Gly Arg Thr Lys Met (SEQ. ID No. 78), Met His Ala Arg Ser Thr Gly Ser Tyr Ser Leu Val Thr Gln Gln Pro Leu Gly Gly Lys Ala Gln Phe Gly (SEQ. ID No. 79)and Gly Gln Arg Phe Gly Glu Met Glu Val Trp Ala Leu Glu Ala Tyr Gly Ala Ala Tyr Thr Leu Gln Glu Met Leu (SEQ. ID No. 80).

160. The drug of Claim 157 wherein the compound is a derivative of an amino acid sequence selected from the group consisting of Met Thr Asp Asn Gly

Thr Phe Val Ile Asn Gly Thr Glu Arg Val Ile Val Ser Gln Leu His Arg (SEQ. ID No. 66), Ser Pro Gly Val Phe Phe Asp Ser Asp Lys Gly Lys Thr His Ser Ser Gly Lys Val Leu Tyr Asn (SEQ. ID No. 67), Ala Arg Ile Ile Pro Tyr Arg Gly Ser Trp Leu Asp Phe Glu Phe Asp Pro Lys Asp Asn Leu Phe (SEQ. ID No. 68), Val Arg Ile Asp Arg Arg Arg Lys Leu Pro Ala Thr Ile Ile Leu Arg Ala Leu Asn Tyr Thr Thr (SEQ. ID No. 69), Glu Gln Ile Leu Asp Leu Phe Phe Glu Lys Val Ile Phe Glu Ile Arg Asp Asn Lys Leu Gln Met (SEQ. ID No. 70), Ser Trp Leu Asp Phe Glu Phe Asp Pro Lys Asp Asn Leu Phe Val Arg Ile Asp Arg Arg Arg Lys Leu Pro (SEQ. ID No. 71), Val Ser Gln Leu His Arg Ser Pro Gly Val Phe Phe Asp Ser Asp Lys Gly Lys Thr His Ser (SEQ. ID No. 72), Ser Gly Lys Val Leu Tyr Asn Ala Arg Ile Ile Pro Tyr Arg Gly Ser Trp Leu (SEQ. ID No. 73), Leu Asp Phe Glu Phe Asp Pro Lys Asp Asn Leu Phe Val Arg Ile Asp Arg Arg Arg Lys Leu Pro (SEQ. ID No. 74), Ala Thr Ile Ile Leu Arg Ala Leu Asn Tyr Thr Thr Glu Gln Ile Leu Asp Leu Phe Phe Glu Lys Val (SEQ. ID No. 75), Met Tyr Met Leu Lys Leu Asn His Leu Val Asp Asp Lys Met His Ala Arg Ser Thr Gly Ser Tyr Ser Leu Val (SEQ. ID No. 76), Thr Gln Gln Pro Leu Gly Gly Lys Ala Gln Phe Gly Gly Gln Arg Phe Gly Glu Met Glu Val Trp Ala Leu Glu (SEQ. ID No. 77), Ala Tyr Gly Ala Ala Tyr Thr Leu Gln Glu Met Leu Thr Val Lys Ser Asp Asp Val Asn Gly Arg Thr Lys Met (SEQ. ID No. 78), Met His Ala Arg Ser Thr Gly Ser Tyr Ser Leu Val Thr Gln Gln Pro Leu Gly Gly Lys Ala Gln Phe Gly (SEQ. ID No. 79)and Gly Gln Arg Phe Gly Glu Met Glu Val Trp Ala Leu Glu Ala Tyr Gly Ala Ala Tyr Thr Leu Gln Glu Met Leu (SEQ. ID No. 80).

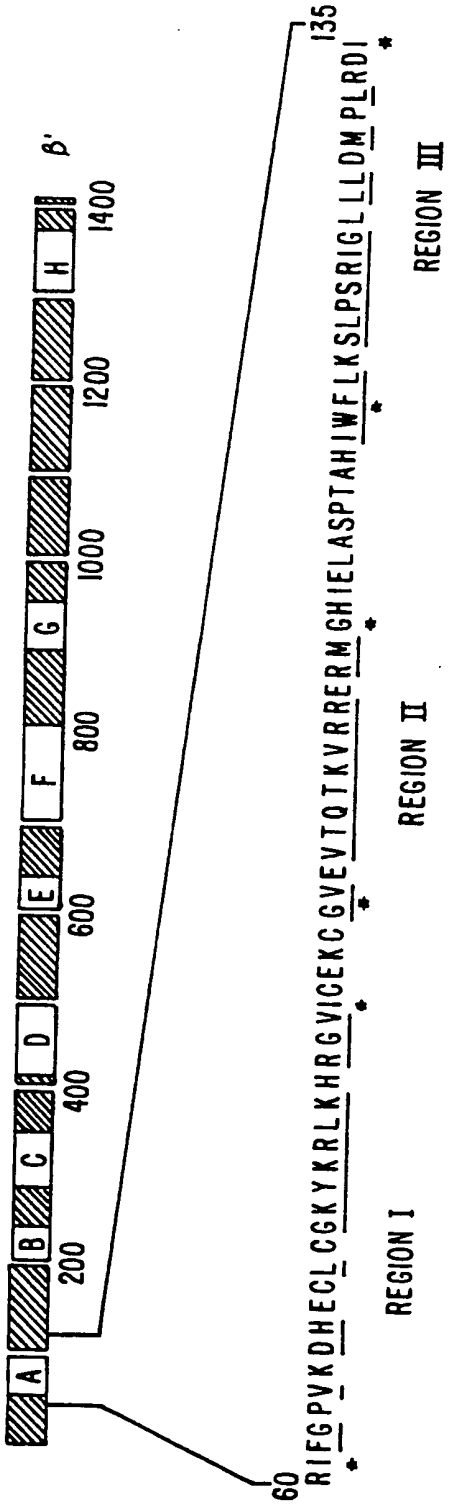

A
B
C
D
E
F
G
H
β'
200
400
600
800
1000
1200
1400
60
135
RIFGPVKDHECLCGKYKRLKHRGVICEKCGVEVTQTKVRRERMGHIELASPTAHIWFLKSLPSRIGLLLDMPLRDI
REGION I
REGION II
REGION III

FIG. 1.

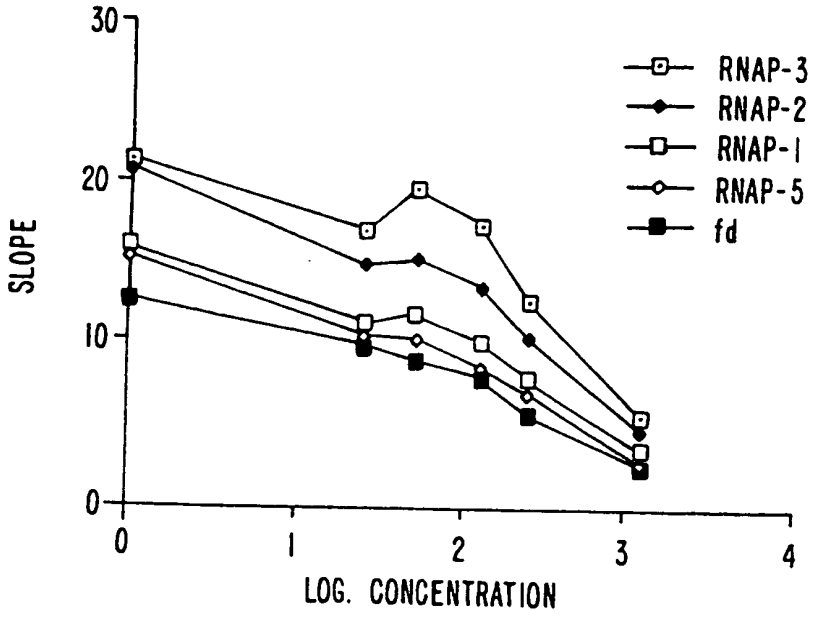

30
20
10
0
SLOPE
0
1
2
3
4
LOG. CONCENTRATION
RNAP-3
RNAP-2
RNAP-1
RNAP-5
fd

FIG. 2A.

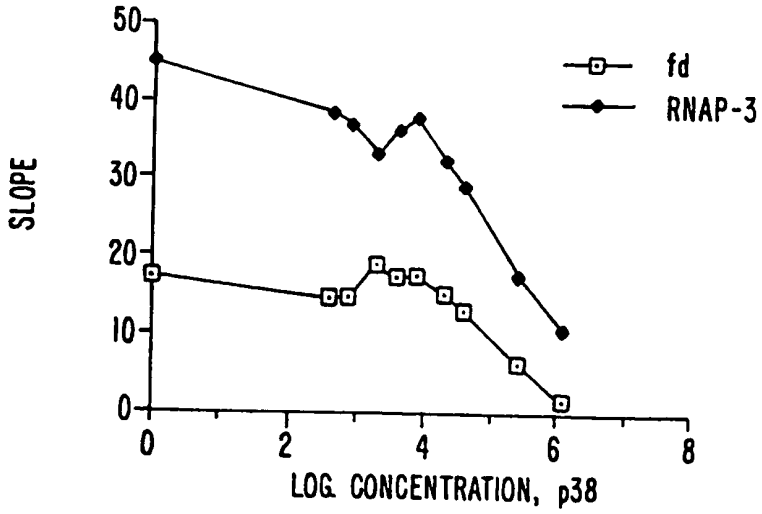
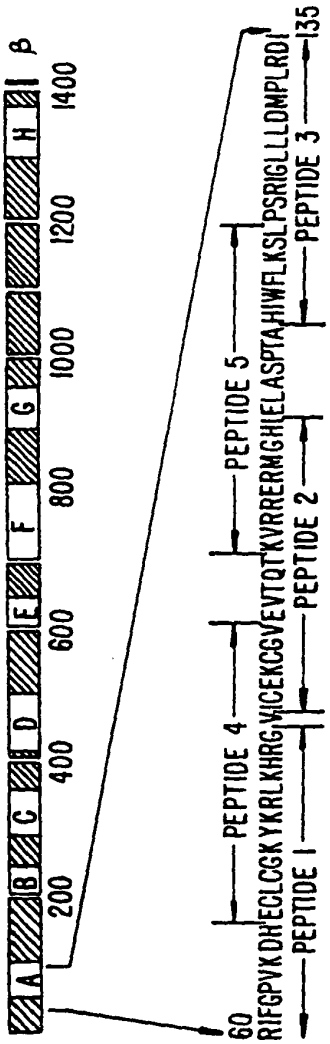

50
40
30
20
10
0
SLOPE
0
2
4
6
8
LOG. CONCENTRATION, p38
fd
RNAP-3

FIG. 2B.



PEPTIDE 1: ARIFGPVKDHEGLCGKYKRLKHRG

PEPTIDE 2: ICEKCGVEVTQTKVRRERMGHI

PEPTIDE 3: CHIWFLKSLPSRIGLLLDMPLRDIE

PEPTIDE 4: ECLCGKYKRLKHRGVICEKCGV

PEPTIDE 5: CKVRRERMGHIELASPTAHIWFLKSL

FIG. 3.

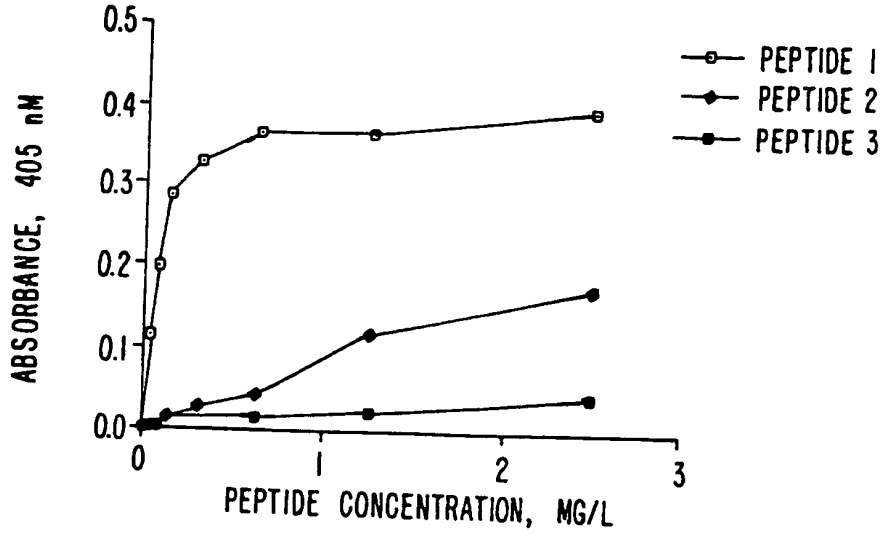

0.5
0.4
0.3
0.2
0.1
0.0
ABSORBANCE, 405 nM
0
1
2
3
PEPTIDE CONCENTRATION, MG/L
PEPTIDE 1
PEPTIDE 2
PEPTIDE 3

FIG. 4A.

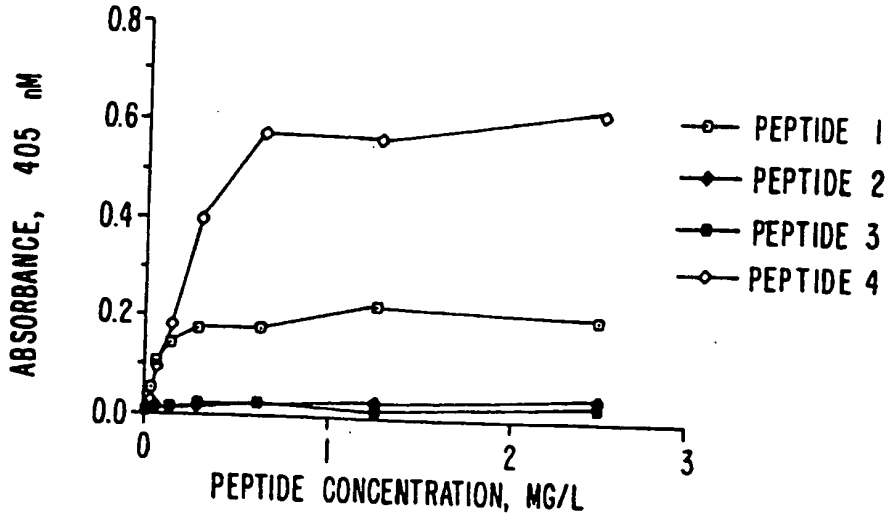

0.8
0.6
0.4
0.2
0.0
ABSORBANCE, 405 nM
0
1
2
3
PEPTIDE CONCENTRATION, MG/L
PEPTIDE 1
PEPTIDE 2
PEPTIDE 3
PEPTIDE 4

FIG. 4B.

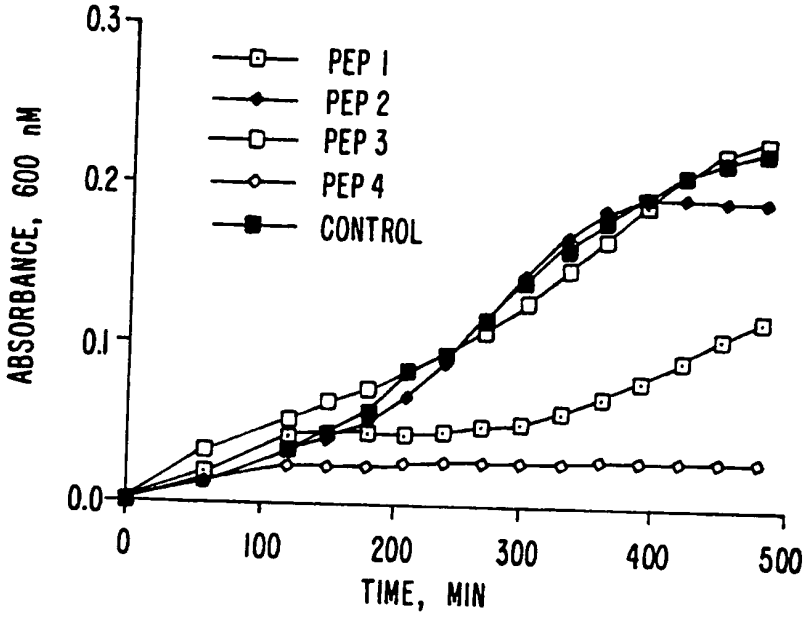

0.3
0.2
0.1
0.0
ABSORBANCE, 600 nM
PEP 1
PEP 2
PEP 3
PEP 4
CONTROL
0
100
200
300
400
500
TIME, MIN

FIG. 5.

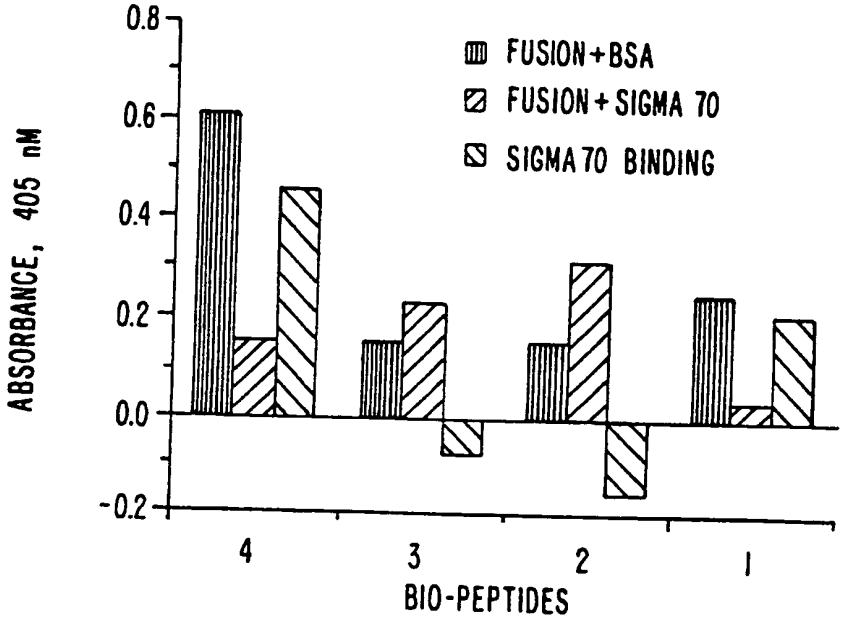

0.8
0.6
0.4
0.2
0.0
-0.2
ABSORBANCE, 405 nM
FUSION+BSA
FUSION+SIGMA 70
SIGMA 70 BINDING
4
3
2
1
BIO-PEPTIDES

FIG. 6.

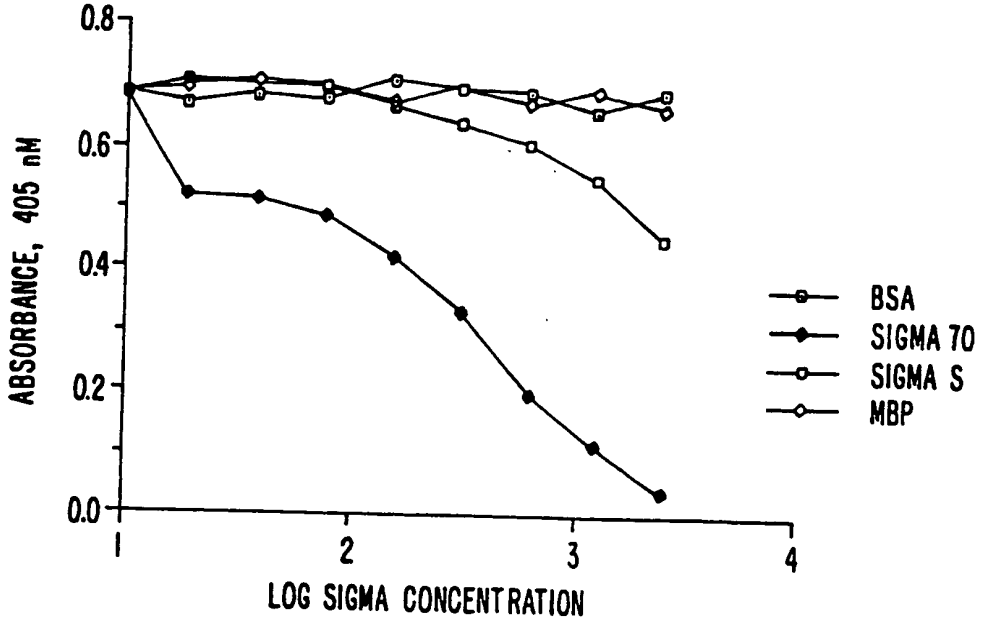

0.8
0.6
0.4
0.2
0.0
ABSORBANCE, 405 nM
1
2
3
4
LOG SIGMA CONCENTRATION
BSA
SIGMA 70
SIGMA S
MBP

FIG. 7.

INTERNATIONAL SEARCH REPORT

International application No.
PCT/US99/04351

A. CLASSIFICATION OF SUBJECT MATTER
IPC(6) : A61K 38/00
US CL : 530/324, 325, 326, 327, 328; 424/158.1
According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)
U.S. : 530/324, 325, 326, 327, 328; 424/158.1

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the international search (name of data base and, where practicable, search terms used)
STN
search terms: RNA Polymerase, inhibitor, antagonist, sub-unit

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
A	HANCOCK R.F.W. "Antibacterial peptides and the outer membranes of Gram-Negative Bacilli" J. Med. Microbiol. 1997. Vol 46. pages 1-3.	NONE
A	WANG et al. "Determinants for Escherichia coli RNA Polymerase Assembly within the Beta Subunit" 1997. Vol. 270. pages 648-662.	
A	LUO et al. " Molecular Anatomy of the Beta' subunit of the E. coli RNA polymerase: Identification of Regions involved in polymerase assembly" 1996. Genes to Cells. Vol. 1, pages 819-827.	

☐ Further documents are listed in the continuation of Box C. ☐ See patent family annex.

* Special categories of cited documents:

"A" document defining the general state of the art which is not considered to be of particular relevance

"E" earlier document published on or after the international filing date

"L" document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)

"O" document referring to an oral disclosure, use, exhibition or other means

"P" document published prior to the international filing date but later than the priority date claimed

"T" later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention

"X" document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone

"Y" document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art

"&" document member of the same patent family

Date of the actual completion of the international search
24 MAY 1999

Date of mailing of the international search report
02 JUL 1999

Name and mailing address of the ISA/US
Commissioner of Patents and Trademarks
Box PCT
Washington, D.C. 20231
Facsimile No. (703) 305-3230

Authorized officer
PATRICK R. DELANEY
Telephone No. (703) 308-0196

Form PCT/ISA/210 (second sheet)(July 1992)*